PATENT COOPERATION TREATY

From the INTERNATIONAL BUREAU

PCT

NOTIFICATION OF ELECTION

(PCT Rule 61.2)

To:

Assistant Commissioner for Patents
United States Patent and Trademark Office
Box PCT
Washington, D.C.20231
ÉTATS-UNIS D'AMÉRIQUE

in its capacity as elected Office

Date of mailing (day/month/year) 04 November 1999 (04.11.99)	
International application No. PCT/GB99/00308	**Applicant's or agent's file reference** JEC/BP5748488
International filing date (day/month/year) 29 January 1999 (29.01.99)	**Priority date** (day/month/year) 29 January 1998 (29.01.98)
Applicant KEITH, William, Nicol	

1. The designated Office is hereby notified of its election made:

[X] in the demand filed with the International Preliminary Examining Authority on:

25 August 1999 (25.08.99)

[ ] in a notice effecting later election filed with the International Bureau on:

2. The election [ ] was

[X] was not

made before the expiration of 19 months from the priority date or, where Rule 32 applies, within the time limit under Rule 32.2(b).

The International Bureau of WIPO
34, chemin des Colombettes
1211 Geneva 20, Switzerland

Facsimile No.: (41-22) 740.14.35

Authorized officer

Lazar Joseph Panakal

Telephone No.: (41-22) 338.83.38

PATENT COOPERATION TREATY

From the INTERNATIONAL SEARCHING AUTHORITY

PCT

NOTIFICATION OF TRANSMITTAL OF THE INTERNATIONAL SEARCH REPORT OR THE DECLARATION

(PCT Rule 44.1)

To:
MEWBURN ELLIS
York House
23 Kingsway
London WC2B 6HP
UNITED KINGDOM

Applicant's or agent's file reference JEC/BP5748488	Date of mailing *(day/month/year)* 19/10/1999 **FOR FURTHER ACTION** See paragraphs 1 and 4 below
International application No. PCT/GB 99/00308	International filing date *(day/month/year)* 29/01/1999
Applicant CANCER RESEARCH CAMPAIGN TECHNOLOGY LIMITED et al.	

1. [X] The applicant is hereby notified that the International Search Report has been established and is transmitted herewith.

Filing of amendments and statement under Article 19:
The applicant is entitled, if he so wishes, to amend the claims of the International Application (see Rule 46):

When? The time limit for filing such amendments is normally 2 months from the date of transmittal of the International Search Report; however, for more details, see the notes on the accompanying sheet.

Where? Directly to the International Bureau of WIPO
34, chemin des Colombettes
1211 Geneva 20, Switzerland
Fascimile No.: (41-22) 740.14.35

For more detailed instructions, see the notes on the accompanying sheet.

2. [ ] The applicant is hereby notified that no International Search Report will be established and that the declaration under Article 17(2)(a) to that effect is transmitted herewith.

3. [ ] **With regard to the protest** against payment of (an) additional fee(s) under Rule 40.2, the applicant is notified that:

[ ] the protest together with the decision thereon has been transmitted to the International Bureau together with the applicant's request to forward the texts of both the protest and the decision thereon to the designated Offices.

[ ] no decision has been made yet on the protest; the applicant will be notified as soon as a decision is made.

4. **Further action(s):** The applicant is reminded of the following:

Shortly after **18 months** from the priority date, the international application will be published by the International Bureau. If the applicant wishes to avoid or postpone publication, a notice of withdrawal of the international application, or of the priority claim, must reach the International Bureau as provided in Rules 90*bis*.1 and 90*bis*.3, respectively, before the completion of the technical preparations for international publication.

Within **19 months** from the priority date, a demand for international preliminary examination must be filed if the applicant wishes to postpone the entry into the national phase until 30 months from the priority date (in some Offices even later).

Within **20 months** from the priority date, the applicant must perform the prescribed acts for entry into the national phase before all designated Offices which have not been elected in the demand or in a later election within 19 months from the priority date or could not be elected because they are not bound by Chapter II.

Name and mailing address of the International Searching Authority European Patent Office, P.B. 5818 Patentlaan 2 NL-2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Authorized officer Andria Overbeeke-Siepkes

NOTES TO FORM PCT/ISA/220

These Notes are intended to give the basic instructions concerning the filing of amendments under article 19. The Notes are based on the requirements of the Patent Cooperation Treaty, the Regulations and the Administrative Instructions under that Treaty. In case of discrepancy between these Notes and those requirements, the latter are applicable. For more detailed information, see also the PCT Applicant's Guide, a publication of WIPO.

In these Notes, "Article", "Rule", and "Section" refer to the provisions of the PCT, the PCT Regulations and the PCT Administrative Instructions, respectively.

INSTRUCTIONS CONCERNING AMENDMENTS UNDER ARTICLE 19

The applicant has, after having received the international search report, one opportunity to amend the claims of the international application. It should however be emphasized that, since all parts of the international application (claims, description and drawings) may be amended during the international preliminary examination procedure, there is usually no need to file amendments of the claims under Article 19 except where, e.g. the applicant wants the latter to be published for the purposes of provisional protection or has another reason for amending the claims before international publication. Furthermore, it should be emphasized that provisional protection is available in some States only.

What parts of the international application may be amended?

Under Article 19, only the claims may be amended.

During the international phase, the claims may also be amended (or further amended) under Article 34 before the International Preliminary Examining Authority. The description and drawings may only be amended under Article 34 before the International Examining Authority.

Upon entry into the national phase, all parts of the international application may be amended under Article 28 or, where applicable, Article 41.

When?

Within 2 months from the date of transmittal of the international search report or 16 months from the priority date, whichever time limit expires later. It should be noted, however, that the amendments will be considered as having been received on time if they are received by the International Bureau after the expiration of the applicable time limit but before the completion of the technical preparations for international publication (Rule 46.1).

Where not to file the amendments?

The amendments may only be filed with the International Bureau and not with the receiving Office or the International Searching Authority (Rule 46.2).

Where a demand for international preliminary examination has been/is filed, see below.

How?

Either by cancelling one or more entire claims, by adding one or more new claims or by amending the text of one or more of the claims as filed.

A replacement sheet must be submitted for each sheet of the claims which, on account of an amendment or amendments, differs from the sheet originally filed.

All the claims appearing on a replacement sheet must be numbered in Arabic numerals. Where a claim is cancelled, no renumbering of the other claims is required. In all cases where claims are renumbered, they must be renumbered consecutively (Administrative Instructions, Section 205(b)).

The amendments must be made in the language in which the international application is to be published.

What documents must/may accompany the amendments?

Letter (Section 205(b)):

The amendments must be submitted with a letter.

The letter will not be published with the international application and the amended claims. It should not be confused with the "Statement under Article 19(1)" (see below, under "Statement under Article 19(1)").

The letter must be in English or French, at the choice of the applicant. However, if the language of the international application is English, the letter must be in English; if the language of the international application is French, the letter must be in French.

The letter must indicate the differences between the claims as filed and the claims as amended. It must, in particular, indicate, in connection with each claim appearing in the international application (it being understood that identical indications concerning several claims may be grouped),whether

(i) the claim is unchanged;

(ii) the claim is cancelled;

(iii) the claim is new;

(iv) the claim replaces one or more claims as filed;

(v) the claim is the result of the division of a claim as filed.

The following examples illustrate the manner in which amendments must be explained in the accompanying letter:

1. [Where originally there were 48 claims and after amendment of some claims there are 51]:
"Claims 1 to 29, 31, 32, 34, 35, 37 to 48 replaced by amended claims bearing the same numbers; claims 30, 33 and 36 unchanged; new claims 49 to 51 added."

2. [Where originally there were 15 claims and after amendment of all claims there are 11]:
"Claims 1 to 15 replaced by amended claims 1 to 11."

3. [Where originally there were 14 claims and the amendments consist in cancelling some claims and in adding new claims]:
"Claims 1 to 6 and 14 unchanged; claims 7 to 13 cancelled; new claims 15, 16 and 17 added." or
"Claims 7 to 13 cancelled; new claims 15, 16 and 17 added; all other claims unchanged."

4. [Where various kinds of amendments are made]:
"Claims 1-10 unchanged; claims 11 to 13, 18 and 19 cancelled; claims 14, 15 and 16 replaced by amended claim 14; claim 17 subdivided into amended claims 15, 16 and 17; new claims 20 and 21 added."

"Statement under article 19(1)" (Rule 46.4)

The amendments may be accompanied by a statement explaining the amendments and indicating any impact that such amendments might have on the description and the drawings (which cannot be amended under Article 19(1)).

The statement will be published with the international application and the amended claims.

It must be in the language in which the international application is to be published.

It must be brief, not exceeding 500 words if in English or if translated into English.

It should not be confused with and does not replace the letter indicating the differences between the claims as filed and as amended. It must be filed on a separate sheet and must be identified as such by a heading, preferably by using the words "Statement under Article 19(1)."

It may not contain any disparaging comments on the international search report or the relevance of citations contained in that report. Reference to citations, relevant to a given claim, contained in the international search report may be made only in connection with an amendment of that claim.

Consequence if a demand for international preliminary examination has already been filed

If, at the time of filing any amendments and any accompanying statement, under Article 19, a demand for international preliminary examination has already been submitted, the applicant must preferably, at the time of filing the amendments (and any statement) with the International Bureau, also file with the International Preliminary Examining Authority a copy of such amendments (and of any statement) and, where required, a translation of such amendments for the procedure before that Authority (see Rules 55.3(a) and 62.2, first sentence). For further information, see the Notes to the demand form (PCT/IPEA/401).

Consequence with regard to translation of the international application for entry into the national phase

The applicant's attention is drawn to the fact that, upon entry into the national phase, a translation of the claims as amended under Article 19 may have to be furnished to the designated/elected Offices, instead of, or in addition to, the translation of the claims as filed.

For further details on the requirements of each designated/elected Office, see Volume II of the PCT Applicant's Guide.

PATENT COOPERATION TREATY

PCT

INTERNATIONAL SEARCH REPORT

(PCT Article 18 and Rules 43 and 44)

Applicant's or agent's file reference	FOR FURTHER ACTION	see Notification of Transmittal of International Search Report (Form PCT/ISA/220) as well as, where applicable, item 5 below.
JEC/BP5748488		
International application No.	International filing date *(day/month/year)*	(Earliest) Priority Date *(day/month/year)*
PCT/GB 99/00308	29/01/1999	29/01/1998
Applicant		
CANCER RESEARCH CAMPAIGN TECHNOLOGY LIMITED et al.		

This International Search Report has been prepared by this International Searching Authority and is transmitted to the applicant according to Article 18. A copy is being transmitted to the International Bureau.

This International Search Report consists of a total of 7 sheets.

[X] It is also accompanied by a copy of each prior art document cited in this report.

1. **Basis of the report**

   a. With regard to the **language,** the international search was carried out on the basis of the international application in the language in which it was filed, unless otherwise indicated under this item.

   [ ] the international search was carried out on the basis of a translation of the international application furnished to this Authority (Rule 23.1(b)).

   b. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international search was carried out on the basis of the sequence listing :

   [ ] contained in the international application in written form.

   [ ] filed together with the international application in computer readable form.

   [X] furnished subsequently to this Authority in written form.

   [X] furnished subsequently to this Authority in computer readble form.

   [X] the statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

   [X] the statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished

2. [X] **Certain claims were found unsearchable** (See Box I).

3. [X] **Unity of invention is lacking** (see Box II).

4. With regard to the **title,**

   [ ] the text is approved as submitted by the applicant.

   [X] the text has been established by this Authority to read as follows:

   PROMOTER REGIONS OF THE MOUSE AND HUMAN TELOMERASE RNA COMPONENT GENES

5. With regard to the **abstract,**

   [X] the text is approved as submitted by the applicant.

   [ ] the text has been established, according to Rule 38.2(b), by this Authority as it appears in Box III. The applicant may, within one month from the date of mailing of this international search report, submit comments to this Authority.

6. The figure of the **drawings** to be published with the abstract is Figure No. 3a, 3b

   [ ] as suggested by the applicant.

   [X] because the applicant failed to suggest a figure.

   [ ] because this figure better characterizes the invention.

   [ ] None of the figures.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/GB 99/00308

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

Although claims 44 and 45 are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

2. [X] Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:

see FURTHER INFORMATION sheet PCT/ISA/210

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

As a result of the prior review under R. 40.2(e) PCT, no additional fees are to be refunded.

1. [X] As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. [ ] As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[X] The additional search fees were accompanied by the applicant's protest.

[ ] No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. Claims: 2-7 (all totally) and claims 1,14,17-18, 37-38 (all partially)

   A nucleic acid comprising a promoter region of the human telomerase RNA gene, constructs, vectors and host cells containing it.

2. Claims: 8-13 (all totally) and claims 1,14,17-18, 37-38 (all partially)

   A nucleic acid comprising a promoter region of the mouse telomerase RNA gene, constructs, vectors and host cells containing it.

3. Claims: 16 (totally) and claims 15,17-22 (all partially)

   A nucleic acid construct comprising a telomerase RNA gene promoter linked to a cytotoxin, and its use in the treatment of cancer.

4. Claims: 23-36 (all totally) and claims 15, 17-22 (all partially)

   A method for screening for a substance being a modulator of the promoter of a telomerase RNA gene, the substances identified and their use in the treatment of cancer or the activation of telomerase.

5. Claims: 39-45

   A system for controlling neoplasia.

Continuation of Box I.2

Present claims 26, 27 and 32 relate to a substance defined by reference to a desirable characteristic or property, namely its ability to modulate the activity of a TR promoter.
The claims cover all substances having this characteristic or property, whereas the application provides support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT for only a very limited number of such substances. In the present case, the claims so lack support, and the application so lacks disclosure, that a meaningful search over the whole of the claimed scope is impossible. Independent of the above reasoning, the claims also lack clarity (Article 6 PCT). An attempt is made to define the substance by reference to a result to be achieved. Again, this lack of clarity in the present case is such as to render a meaningful search over the whole of the claimed scope impossible. Consequently, the search has been carried out for those parts of the claims which appear to be clear, supported and disclosed, namely those parts relating to the specific modulators as defined in claims 28, 29 and 33.

The applicant's attention is drawn to the fact that claims, or parts of claims, relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

INTERNATIONAL SEARCH REPORT

[redacted]tional Application No
PCT/GB 99/00308

A. CLASSIFICATION OF SUBJECT MATTER
IPC 6 C12N15/11 C12N9/12 C12N15/85 C12Q1/68

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 6 C12N C12Q

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 96 01835 A (ANDREWS WILLIAM H ;VILLEPONTEAU BRYANT (US); FUNK WALTER (US); FEN) 25 January 1996 (1996-01-25)	1-7,14, 15,17, 18, 23-28, 35,37,38
	page 24, line 13 -page 31, line 19	
Y	page 37, line 16 -page 43, line 13	15-21, 26-28, 30,31, 35,39-45
	page 47, line 15 -page 55, line 19	
A	page 65, line 9 -page 66, line 14	26-38

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[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
5 October 1999	19. 10. 1999

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Andres, S

2

Form PCT/ISA/210 (second sheet) (July 1992)

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	PALMITER R D ET AL: "CELL LINEAGE ABLATION IN TRANSGENIC MICE BY CELL-SPECIFIC EXPRESSION OF A TOXIN GENE" CELL, vol. 50, 31 July 1987 (1987-07-31), pages 435-443, XP002055685 ISSN: 0092-8674 the whole document	15-21
Y	FENG J ET AL: "THE RNA COMPONENT OF HUMAN TELOMERASE" SCIENCE, vol. 269, no. 5228, 1 September 1995 (1995-09-01), pages 1236-1241, XP000645335 ISSN: 0036-8075 cited in the application	26-28, 30,31,35
A	the whole document	1-7
Y	RING, C. ET AL.: "Suicide gene expression induced in tumour cells transduced with recombinant adenoviral, retroviral and plasmid vectors containing the ERBB2 promoter" GENE THERAPY., vol. 3, 1996, pages 1094-1103, XP002117436 ISSN: 0969-7128 the whole document	39-45
X	WO 96 01614 A (ANDREWS WILLIAM H ;VILLEPONTEAU BRYANT (US); FUNK WALTER (US); GRE) 25 January 1996 (1996-01-25) cited in the application page 12, line 28 -page 13, line 25 figures 1,3 example 7	1-8,14, 17,18
X	HINKLEY, C. ET AL.: "The mouse telomerase RNA 5'-end lies just upstream of the telomerase template sequence" NUCLEIC ACIDS RESEARCH., vol. 26, 15 January 1998 (1998-01-15), pages 532-536, XP002106807 OXFORD UNIVERSITY PRESS, SURREY., GB ISSN: 0305-1048 the whole document	1-3,8,9, 14,17,18
X	PARKINSON E K ET AL: "The genetic basis of human keratinocyte immortalisation in squamous cell carcinoma development: the role of telomerase reactivation." EUROPEAN JOURNAL OF CANCER, (1997 APR) 33 (5) 727-34, XP002117437	26,31, 32,34, 36-38
A	page 731, paragraph 2 -page 732	26-38

-/--

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	LANDBERG, G. ET AL.: "Telomerase activity is associated with cell cycle deregulation in human breast cancer" CANCER RESEARCH., vol. 57, 1 February 1997 (1997-02-01), pages 549-554, XP002117438 ISSN: 0008-5472 ---	26-38
A	HART L R : "TISSUE- SPECIFIC PROMOTERS IN TARGETING SYSTEMICALLY DELIVERED GENE-THERAPY" SEMINARS IN ONCOLOGY, (FEB 1996) VOL. 23, NO. 1, PP. 154-158., XP002117439 the whole document ---	39-45
A	WO 95 06486 A (OLDFIELD EDWARD J ;RAM ZVI (US); US HEALTH (US); BLAESE R MICHAEL) 9 March 1995 (1995-03-09) page 14, last paragraph -page 15, line 6 example 1 claims ---	39-45
A	HARLEY, C. & SHERWOOD, S.: "Telomerase, checkpoints and cancer" CANCER SURVEYS, vol. 29, 1997, pages 263-284, XP002117440 ---	
A	GREIDER C W: "TELOMERE LENGTH REGULATION" ANNUAL REVIEW OF BIOCHEMISTRY, vol. 65, 1996, pages 337-365, XP002056801 ---	
P,X	ZHAO, J.-Q. ET AL.: "Cloning and characterization of human and mouse telomerase RNA gene promoter sequences." ONCOGENE, vol. 16, 1998, pages 1345-1350, XP002106808 ISSN: 0950-9232 the whole document ---	1-15,17, 18
P,X	WO 98 11207 A (VILLEPONTEAU BRYANT ;HARLEY CALVIN (US); GERON CORP (US)) 19 March 1998 (1998-03-19) page 23, line 25 -page 32, line 13 page 33, line 27 -page 38 -----	1-7,15, 17-26, 30-32, 34-38

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/GB 99/00308

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9601835 A	25-01-1996	US 5583016 A	10-12-1996
		US 5776679 A	07-07-1998
		AU 696702 B	17-09-1998
		AU 2964795 A	09-02-1996
		AU 3095095 A	09-02-1996
		AU 9712998 A	18-03-1999
		BG 101103 A	31-10-1997
		BR 9508254 A	23-12-1997
		CA 2194393 A	25-01-1996
		CN 1158617 A	03-09-1997
		CN 1168696 A	24-12-1997
		CZ 9700034 A	15-10-1997
		EP 0778842 A	18-06-1997
		EP 0793719 A	10-09-1997
		FI 970026 A	03-03-1997
		HU 78054 A	28-07-1999
		JP 10505488 T	02-06-1998
		JP 10507067 T	14-07-1998
		NO 970041 A	06-03-1997
		NZ 289720 A	29-03-1999
		PL 318169 A	26-05-1997
		WO 9601614 A	25-01-1998
		US 5876979 A	02-03-1999
		US 5837857 A	17-11-1998
WO 9601614 A	25-01-1996	US 5583016 A	10-12-1996
		US 5876979 A	02-03-1999
		AU 696702 B	17-09-1998
		AU 2964795 A	09-02-1996
		AU 3095095 A	09-02-1996
		AU 9712998 A	18-03-1999
		BG 101103 A	31-10-1997
		BR 9508254 A	23-12-1997
		CA 2194393 A	25-01-1996
		CN 1158617 A	03-09-1997
		CN 1168696 A	24-12-1997
		CZ 9700034 A	15-10-1997
		EP 0778842 A	18-06-1997
		EP 0793719 A	10-09-1997
		FI 970026 A	03-03-1997
		HU 78054 A	28-07-1999
		JP 10505488 T	02-06-1998
		JP 10507067 T	14-07-1998
		NO 970041 A	06-03-1997
		NZ 289720 A	29-03-1999
		PL 318169 A	26-05-1997
		WO 9601835 A	25-01-1996
		US 5776679 A	07-07-1999
		US 5837857 A	17-11-1998
WO 9506486 A	09-03-1995	CA 2169557 A	09-03-1995
		EP 0716614 A	19-06-1996
		JP 9505557 T	03-06-1997
WO 9811207 A	19-03-1998	AU 4351997 A	02-04-1998

PCT

REQUEST

The undersigned requests that the present international application be processed according to the Patent Cooperation Treaty

For receiving Office use only

International Application No.

International Filing Date

Name of receiving Office and "PCT International Application"

Applicant's or agent's file reference *(if desired) (12 characters maximum)* JEC/BP5748488

Box No. I TITLE OF INVENTION A GENE PROMOTER

Box No. II APPLICANT

Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country. The country of the address indicated in this Box is the applicant's State (that is, country) of residence if no State of residence is indicated below.)*

CANCER RESEARCH CAMPAIGN TECHNOLOGY LIMITED
Cambridge House
6-10 Cambridge Terrace
Regent's Park
LONDON
NW1 4JL
GB

☐ This person is also inventor.

Telephone No.

Facsimile No.

Teleprinter No.

State *(that is, country) of nationality:* GB

State *(that is, country) of residence:* GB

This person is applicant for the purposes of: ☐ all designated States ☒ all designated States except the United States of America ☐ the United States of America only ☐ the States indicated in the Supplemental Box

Box No. III FURTHER APPLICANT(S) AND/OR (FURTHER) INVENTOR(S)

Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country. The country of the address indicated in this Box is the applicant's State (that is, country) of residence if no State of residence is indicated below.)*

KEITH William Nicol
CRC Department of Medical Oncology
University of Glasgow
CRC Beatson Laboratories
Garscube Estate
Switchback Road
Glasgow
G61 1BD
GB

This person is:

☐ applicant only

☒ applicant and inventor

☐ inventor only *(if this check-box is marked, do not fill in below.)*

State *(that is, country) of nationality:* GB

State *(that is, country) of residence:* GB

This person is applicant for the purposes of: ☐ all designated States ☐ all designated States except the United States of America ☒ the United States of America only ☐ the States indicated in the Supplemental Box

☐ Further applicants and/or (further) inventors are indicated on a continuation sheet.

Box No. IV AGENT OR COMMON REPRESENTATIVE; OR ADDRESS FOR CORRESPONDENCE

The person identified below is hereby/has been appointed to act on behalf of the applicant(s) before the competent International Authorities as: ☒ agent ☐ common representative

Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*

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☐ Mark this check-box where no agent or common representative is/has been appointed and the space above is used instead to indicate a special address to which correspondence should be sent.

Box No. V DESIGNATION OF STATES

The following designations are hereby made under Rule 4.9(a) *(mark the applicable check-boxes; at least one must be marked)*:

Regional patent

- [X] AP — ARIPO Patent: GH Ghana, GM Gambia, KE Kenya, LS Lesotho, MW Malawi, SD Sudan, SZ Swaziland, UG Uganda, ZW Zimbabwe, and any other State which is a Contracting State of the Harare Protocol and of the PCT
- [X] EA — Eurasian Patent: AM Armenia, AZ Azerbaijan, BY Belarus, KG Kyrgyzstan, KZ Kazakstan, MD Republic of Moldova, RU Russian Federation, TJ Tajikistan, TM Turkmenistan, and any other State which is a Contracting State of the Eurasian Patent Convention and of the PCT
- [X] EP — European Patent: AT Austria, BE Belgium, CH and LI Switzerland and Liechtenstein, CY Cyprus, DE Germany, DK Denmark, ES Spain, FI Finland, FR France, GB United Kingdom, GR Greece, IE Ireland, IT Italy, LU Luxembourg, MC Monaco, NL Netherlands, PT Portugal, SE Sweden, and any other State which is a Contracting State of the European Patent Convention and of the PCT
- [X] OA — OAPI Patent: BF Burkina Faso, BJ Benin, CF Central African Republic, CG Congo, CI Côte d'Ivoire, CM Cameroon, GA Gabon, GN Guinea, GW Guinea-Bissau, ML Mali, MR Mauritania, NE Niger, SN Senegal, TD Chad, TG Togo, and any other State which is a member State of OAPI and a Contracting State of the PCT *(if other kind of protection or treatment desired, specify on dotted line)*

.....

National Patent *(if other kind of protection desired, specify on dotted line)*:

- [X] AL Albania
- [X] AM Armenia
- [X] AT Austria
- [X] AU Australia
- [X] AZ Azerbaijan
- [X] BA Bosnia & Herzegovina
- [X] BB Barbados
- [X] BG Bulgaria
- [X] BR Brazil
- [X] BY Belarus
- [X] CA Canada
- [X] CH and LI Switzerland and Liechtenstein
- [X] CN China
- [X] CU Cuba
- [X] CZ Czech Republic
- [X] DE Germany
- [X] DK Denmark
- [X] EE Estonia
- [X] ES Spain
- [X] FI Finland
- [X] GB United Kingdom
- [X] GE Georgia
- [X] GH Ghana
- [X] GM Gambia
- [X] HR Croatia
- [X] HU Hungary
- [X] ID Indonesia
- [X] IL Israel
- [X] IS Iceland
- [X] JP Japan
- [X] KE Kenya
- [X] KG Kyrgyzstan
- [X] KP Democratic People's Republic of Korea
- [X] KR Republic of Korea
- [X] KZ Kazakstan
- [X] LC St Lucia
- [X] LK Sri Lanka
- [X] LR Liberia
- [X] LS Lesotho
- [X] LT Lithuania
- [X] LU Luxembourg
- [X] LV Latvia
- [X] MD Republic of Moldova
- [X] MG Madagascar
- [X] MK The former Yugoslav Republic of Macedonia
- [X] MN Mongolia
- [X] MW Malawi
- [X] MX Mexico
- [X] NO Norway
- [X] NZ New Zealand
- [X] PL Poland
- [X] PT Portugal
- [X] RO Romania
- [X] RU Russian Federation
- [X] SD Sudan
- [X] SE Sweden
- [X] SG Singapore
- [X] SI Slovenia
- [X] SK Slovakia
- [X] SL Sierra Leone
- [X] TJ Tajikistan
- [X] TM Turkmenistan
- [X] TR Turkey
- [X] TT Trinidad and Tobago
- [X] UA Ukraine
- [X] UG Uganda
- [X] US United States of America
- [X] UZ Uzbekistan
- [X] VN Viet Nam
- [X] YU Yugoslavia
- [X] ZW Zimbabwe

Check-boxes reserved for designating States (for the purposes of a national patent) which have become party to the PCT after issuance of this sheet:

- [X] GD Grenada
- [X] IN India
- [X] Any other state which is party to the PCT

Precautionary Designation Statement: In addition to the designations made above, the applicant also makes under Rule 4.9(b) all designations which would be permitted under the PCT except any designation(s) indicated in the Supplemental Box as being excluded from the scope of this statement.

The applicant declares that those additional designations are subject to confirmation and that any designation which is not confirmed before the expiration of 15 months from the priority date is to be regarded as withdrawn by the applicant at the expiration of that time limit.

(Confirmation of a designation consists of the filing of a notice specifying that designation and the payment of the designation and confirmation fees. Confirmation must reach the receiving Office within the 15-month time limit.)

Supplemental Box *If the Supplemental Box is not used, this sheet need not be included in the request.*

Use this box in the following cases:

I. If, in any of the Boxes, the space is insufficient to furnish all the information:	*In such case, write "Continuation of Box No. ..." (indicate the number of the Box) and furnish the information in the same manner as required according to the captions of the Box in which the space was insufficient;*
in particular:	
(i) if more than two persons are involved as applicants and/or inventors and no "continuation sheet" is available:	*in such case, write "Continuation of Box III" and indicate for each additional person the same type of information as required in Box No. III. The country of the address indicated in this box is the applicant's state (that is, country) of residence if no state of residence is indicated below;*
(ii) if, in Box No. II or in any of the sub-boxes of Box No. III, the indication "the States indicated in the Supplemental Box" is checked:	*in such case write "Continuation of Box No. II" or "Continuation of Box No. III" or "Continuation of Boxes No. II and No. III" (as the case may be), indicate the name of the applicant(s) involved and next to (each) such name, the State(s) (and/or, where applicable, ARIPO, Eurasian, European or OAPI patent) for the purposes of which the named person is applicant;*
(iii) if, in Box No. II or in any of the sub-boxes of Box No. III, the inventor or the inventor/applicant is not inventor for the purposes of all designated States or for the purposes of the United States of America:	*in such case, write "Continuation of Box No. II" or "Continuation of Box No. III" or "Continuation of Boxes No. II and No. III" (as the case may be), indicate the name of the inventor(s) and, next to (each) such name, the State(s) (and/or, where applicable, ARIPO, Eurasian, European or OAPI patent) for the purposes of which the named person is inventor;*
(iv) if, in addition to the agent(s) indicated in Box No. IV, there are further agents:	*in such case, write "Continuation of Box No. IV" and indicate for each further agent the same type of information as required in Box No. IV;*
(v) if, in Box No. V, the name of any State (or OAPI) is accompanied by the indication "patent of addition," or "certificate of addition," or if, in Box No. V, the name of the United States of America is accompanied by an indication "Continuation" or "Continuation-in-part":	*in such case, write "Continuation of Box No. V" and the name of each State involved (or OAPI), and after the name of each such State (or OAPI), the number of the parent title or parent application and the date of grant of the parent title or filing of the parent application;*
(vi) if in Box No. VI there are more than three earlier applications whose priority is claimed:	*in such case, write "Continuation of Box No. VI" and indicate for each additional earlier application the same type of information as required in Box No. VI.*
(vii) if, in Box No. VI, the earlier application is an ARIPO application:	*in such case, write "Continuation of Box No. VI", specify the number of the item corresponding to that earlier application and indicate at least one country party to the Paris Convention for the Protection of Industrial Property for which that earlier application was filed.*
2. If, with regard to the precautionary designation statement contained in Box No. V, the applicant wishes to exclude any State(s) from the scope of that statement:	*in such case, write "Designation(s) excluded from precautionary designation statement" and indicate the name or two-letter code of each state so excluded.*
3. If the applicant claims, in respect of any designated Office, the benefits of provisions of the national law concerning non-prejudicial disclosures or exceptions to lack of novelty:	*in such case, write "Statement Concerning Non-Prejudicial Disclosures or Exceptions to Lack of Novelty" and furnish that statement below.*

Continuation of Box IV

ARMITAGE, IAN M.
BRASNETT, ADRIAN H.
BREWSTER, ANDREA R.
CALDERBANK, T. ROGER
CARTER, STEVEN J.
COLEIRO, RAYMOND
CRIPPS, JOANNA E.
FORD, MICHAEL F.
GURA, H. ALAN
HACKNEY, NIGEL J.
HAMILTON, ALISTAIR
HARRISON, DAVID C.
KIDDLE, SIMON J.
KREMER, SIMON M.
LINN, S. JONATHAN
LYONS, JUNE, M.
NICHOLLS, KATHRYN M.
O'BRIEN, CAROLINE J.
PAGET, HUGH C.E.
SANDERSON, MICHAEL J.
STONER, G. PATRICK
STUART, IAN
WALTON, SEÁN M.
WATKIN, TIMOTHY L.

Box No. VI **PRIORITY CLAIM** ☐ Further priority claims are indicated in the Supplemental Box

Filing date of earlier application (day/month/year)	Number of earlier application	Where earlier application is: national application: country	regional application: * regional Office	international application: receiving Office
item (1) 29 January 1998	9801902.9	GB		
item (2)				
item (3)				

☒ The receiving Office is requested to prepare and transmit to the International Bureau a certified copy of the earlier application(s) *(only if the earlier application was filed with the Office which for the purposes of the present international application is the receiving Office) identified above as item(s):* (1)

** Where the earlier application is an ARIPO application, it is mandatory to indicate in the supplemental box at least one country party to the Paris Convention fo the Protection of Industrial Property for which that earlier application was filed (Rule 4.10(b)(ii)). See Supplemental Box.*

Box No. VII **INTERNATIONAL SEARCHING AUTHORITY**

Choice of International Searching Authority (ISA) *(If two or more International Searching Authorities are competent to carry out the international search, indicate the Authority chosen; the two-letter code may be used):*

ISA /

Request to use results of earlier search; reference to that search (if an earlier search has been carried out by or requested from the International Searching Authority):

Date *(day/month/year)* Number Country *(or regional Office)*

Box No. VIII **CHECK LIST; LANGUAGE OF FILING**

This international application contains the following number of sheets	
request	:4
description (excluding sequence listing part)	:71
claims	:7
abstract	:1
drawings	:29
sequence listing part of description	:0
Total number of sheets	:112

This international application is accompanied by the item(s) marked below:

1. ☒ fee calculation sheet
2. ☒ separate signed power of attorney
3. ☒ copy of general power of attorney; reference number, if any:
4. ☐ statement explaining lack of signature
5. 0 priority document(s) *identified in Box No. VI as item*(s):
6. ☐ translation of international application into (language):
7. ☐ separate indications concerning deposited microorganisms or other biological material
8. ☐ nucleotide and/or amino acid sequence listing in computer readable form
9. ☒ other *(specify)*:Form 23/77

Figure of the drawings which should accompany the abstract: 5

Language of filing of the international application: ENGLISH

Box No. IX **SIGNATURE OF APPLICANT OR AGENT**

Next to each signature indicate the name of the person signing and the capacity in which the person signs (if such capacity is not obvious from reading the request).

.....

CRIPPS, JOANNA E. (Appointed Agent)
APPOINTED AGENT

For receiving Office use only

1. Date of actual receipt of the purported international application:	2. Drawings:
3. Corrected date of actual receipt due to later but timely received papers or drawings completing the purported international application:	☐ received:
4. Date of timely receipt of the required corrections under PCT Article 11(2):	☐ not received:
5. International Searching Authority (if two or more are competent: ISA/	6. ☐ Transmittal of search copy delayed until search fee is paid

For International Bureau use only

Date of receipt of the record copy by the International Bureau:

PATENT COOPERATION TREATY

n the
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

To:

CRIPPS, Joanna E. et al.
Mewburn Ellis
York House
23 Kingsway
London WC2B 6HP
GRANDE BRETAGNE

CEIVED
25 APR 2000

PCT

NOTIFICATION OF TRANSMITTAL OF THE INTERNATIONAL PRELIMINARY EXAMINATION REPORT
(PCT Rule 71.1)

Date of mailing *(day/month/year)*	20.04.00
Applicant's or agent's file reference JEC/BP5748488	IMPORTANT NOTIFICATION

International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/GB99/00308	29/01/1999	29/01/1998

Applicant
CANCER RESEARCH CAMPAIGN TECHNOLOGY LIMITED et al.

1. The applicant is hereby notified that this International Preliminary Examining Authority transmits herewith the international preliminary examination report and its annexes, if any, established on the international application.

2. A copy of the report and its annexes, if any, is being transmitted to the International Bureau for communication to all the elected Offices.

3. Where required by any of the elected Offices, the International Bureau will prepare an English translation of the report (but not of any annexes) and will transmit such translation to those Offices.

4. **REMINDER**

The applicant must enter the national phase before each elected Office by performing certain acts (filing translations and paying national fees) within 30 months from the priority date (or later in some Offices) (Article 39(1)) (see also the reminder sent by the International Bureau with Form PCT/IB/301).

Where a translation of the international application must be furnished to an elected Office, that translation must contain a translation of any annexes to the international preliminary examination report. It is the applicant's responsibility to prepare and furnish such translation directly to each elected Office concerned.

For further details on the applicable time limits and requirements of the elected Offices, see Volume II of the PCT Applicant's Guide.

Name and mailing address of the IPEA/	Authorized officer
European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Vullo, C Tel.+49 89 2399-8061

Form PCT/IPEA/416 (July 1992)

PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference	FOR FURTHER ACTION	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
JEC/BP5748488		
International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/GB99/00308	29/01/1999	29/01/1998
International Patent Classification (IPC) or national classification and IPC		
C12N15/11		
Applicant		
CANCER RESEARCH CAMPAIGN TECHNOLOGY LIMITED et al.		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 9 sheets, including this cover sheet.

   ☒ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of 5 sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the report
   - II ☐ Priority
   - III ☐ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV ☒ Lack of unity of invention
   - V ☒ Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations suporting such statement
   - VI ☒ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☒ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
25/08/1999	20.04.00
Name and mailing address of the international preliminary examining authority:	Authorized officer
European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Moonen, P Telephone No. +49 89 2399 8538

Form PCT/IPEA/409 (cover sheet) (January 1994)

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/GB99/00308

I. Basis of the report

1. This report has been drawn on the basis of (*substitute sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to the report since they do not contain amendments.*):

Description, pages:

1-71	as originally filed				

Claims, No.:

1-25	as received on	31/03/2000	with letter of	27/03/2000	

Drawings, sheets:

1/29-29/29	as originally filed

2. The amendments have resulted in the cancellation of:

☐ the description, pages:
☐ the claims, Nos.:
☐ the drawings, sheets:

3. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)):

4. Additional observations, if necessary:

IV. Lack of unity of invention

1. In response to the invitation to restrict or pay additional fees the applicant has:

☐ restricted the claims.

☐ paid additional fees.

☐ paid additional fees under protest.

☒ neither restricted nor paid additional fees.

2. ☐ This Authority found that the requirement of unity of invention is not complied and chose, according to Rule 68.1, not to invite the applicant to restrict or pay additional fees.

3. This Authority considers that the requirement of unity of invention in accordance with Rules 13.1, 13.2 and 13.3 is

   ☐ complied with.

   ☒ not complied with for the following reasons:

   see separate sheet

4. Consequently, the following parts of the international application were the subject of international preliminary examination in establishing this report:

   ☐ all parts.

   ☒ the parts relating to claims Nos. 2-3 completely; 1,4-5,8-9,17-18 partially.

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	2-3 (completely); 1, 4-5, 8-9, 17-18 (partially)
	No:	Claims	
Inventive step (IS)	Yes:	Claims	
	No:	Claims	2-3 (completely); 1, 4-5, 8-9, 17-18 (partially)
Industrial applicability (IA)	Yes:	Claims	2-3 (completely); 1, 4-5, 8-9, 17-18 (partially)
	No:	Claims	

2. Citations and explanations

   see separate sheet

VI. Certain documents cited

1. Certain published documents (Rule 70.10)

   and / or

2. Non-written disclosures (Rule 70.9)

   see separate sheet

VIII. Certain observations on the international application

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:

see separate sheet

Reference is made to the following documents, cited in the Search Report:

D1: WO 96 01835 (Geron Corp.)
D2: Nucleic Acids Research **26** (15.01.98) 532-536 (Hinkley et al.)
D3: WO 96 01614 (CSH LAB et al.)
D4: Oncogene **16** (1998) 1345-50; P,X-document
D5: WO 98 11207 (Villeponteau et al.), published 19-03-1998; P,X-document

Introduction

1. The International Search Authority (ISA) identified in the originally filed claims five inventions for the reasons mentioned in the extra sheets attached to Form PCT/ISA/206 sent to the Applicant 14.07.99.

   The Applicant decided to pay all the requested 4 additional search fees under protest with an extensive motivation in the reply letter of 13.08.99.

   The Review Panel of the International Search Authority confirmed the reasons for the request to either restrict the claimed subject-matter or pay additional fees.

   The present International Preliminary Examining Authority issued on 17.12.1999 an invitation to either restrict or pay additional fees. The Applicant's response has been the filing of a new set of claims, still considered not to be unitarily linked and the **first invention** identified in the invitation to restrict or pay is therefore **only** the subject of **further examination** (concerning an isolated promoter region derived of the **human** telomerase RNA (TR) gene promoter, constructs, vectors and host cells containing it): this invention is presently referred to in **claims 2-3** (completely), **and claims 1, 4-5, 8-9, 17-18** (partially, in so far as referring to human sequences only).

Re Item IV

Lack of unity of invention

2. The International Preliminary Examining Authority (IPEA) agreed with the ISA in the identification of the independent inventions in the present application, which

for the present set of claims is as follows:

i. **Claims 2-3** (completely), **and claims 1, 4-5, 8-9, 17-18** (partially): concerning an isolated promoter sequence derived from the **human** TR gene, constructs, vectors and host cells containing it;

ii. **Claims 1, 4-5, 8-9, 17-18** (partially): concerning an isolated promoter sequence derived from **an undefined (eg mouse)** telomerase RNA gene, constructs, vectors and host cells containing it;

iii. **Claim 6-7 and 10-13** (completely), **and claims 8-9, 17-18** (partially): concerning a nucleic acid construct of undefined length comprising a telomerase RNA gene promoter sequence operably linked to a heterologous gene cytotoxin, and its use in the treatment of cancer;

iv. **Claims 14-16**: concerning a method for screening for a substance being a modulator of the promoter of a telomerase RNA gene, the substances identified and their use in the treament of cancer or the activation of telomerase;

v. **Claims 19-25**: concerning a system to control neoplasia or method of treating neoplasia.

3. With respect to the reasons for the observation of non-unity the following is noted:

A single inventive concept is not provided by the feature of an "isolated promoter sequence (initiating transcription of DNA operably linked downstream) derived from the telomerase RNA gene promoter having approx. 505 bp upstream of the transcription start site or a fragment thereof" (the length of the promoter still undefined by the wording having), in particular on the basis of the disclosure of D2: see e.g. page 534 paragraph with the heading "DNA-sequence analysis of **human and mouse telomerase RNA promoter regions**" and Figure 3. The sequence of the mouse telomerase-RNA gene extends 89 bp upstream of the transcription start site indicated in Figure 3 and contains several identified promoter elements and therefore is considered to be prejudicial to the novelty of the subject-matter of claim 1 (although the transcriptional start site is differently identified).

Even if account is taken from the fact that none of the cited prior art documents comprise a (fully) identified **functional** promoter than it is submitted that the skilled person was in the position to identify the functional promoter sequence on

the basis of the separate disclosures of the human and mouse TR gene sequence: see for the human sequence D1, as well as Figure 1B and 7A of D3 with the transcriptional start site at bp 1459 in Fig.1B; for the mouse sequence see D2, as well as Figure 3 of D3 with the presently identified transcriptional start at bp 606. It is noted that transcriptional start sites for both the human and mouse TR RNA genes had already been established (see e.g. Figure 7A of D3) and assays for detecting promoter activity in the 5'-flanking regions were already known (e.g. a firefly luciferase reporter gene). Thus, between the identification of the full human and mouse TR promoter sequences already present in disclosed sequences flanking the 5'-end of known telomerase RNA-ccoding sequences there is no new and inventive link. The system of claim 19 based in part to a claim lacking novelty (e.g. claim 1) is not necessarily linked to the inventive concepts of identified inventions I and ii.
A similar reasoning applies to inventions ii and iv: nucleic acid sequences comprising a TR promoter or a fragment thereof were known (see e.g. D3 as mentioned above) and the concepts of inventions ii and iv are neither linked with each other nor with the finding of the isolated sequences of the full functional human and mouse TR promoter.

Therefore, a common, special technical feature referred to in Rule 13 PCT and the PCT Preliminary Examination Guidelines Ch.III, 7 is missing.

Re Item V

Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

4. The presently examined application refers to the finding of the actual sequence of the human TR gene promoter bases in the complete sequence of the TR gene.

D1-D3 (and D5) are all publications by the same research group or researchers closely working together. D2 is the most recent prior art study for identifying the mouse telomerase RNA 5'-end (just 2 nt from the telomerase-RNA template sequence) and also the 5'-end of the human telomerase 45 nt from the telomerase-RNA template sequence: it does not claim the isolation and identification of the 5'-end flanking region with the functional promoter, only of

related human and mouse telomerase RNA promoter elements in specified regions (page 534 right column), with homology to elements typical of RNA polymerase II mRNA-type promoters. However, the identified TATA and CCAAT boxes (see Figure 3) appear to act as a "minimal promoter"; D2 is considered to be the document providing the skilled person with the teaching for further study of the transcriptional unit flanking the 5'-end comprising e.g. enhancer, CCAAT box, TATA box, Sp1 site etc. (also referred to on page 21 of D1). Furthermore, D1 has referred to the modulation of the transcription of a reporter polynucleotide sequence (e.g. a luciferase gene) operably linked to a transcriptional regulatory sequence (promoter and **upstream** transcription factor binding sites) of a human telomerase RNA component gene in a metabolically active mammalian cell (see paragraph bridging pages 12-13 of D1). Indeed the sequence contains e.g. consensus sequences for e.g. Sp1.

It is therefore considered to have been obvious for the skilled person at the priority date which steps to take in the identification of the transcriptional regulatory sequence, that is the linking of the sequence of the 1459 bases as well as the sequence of about 60 bases containing only the CCAAT and TATA boxes to a reporter polynucleotide sequence in addition to sequences having added bases upstream of said 60 bases in correspondence to the sequence of the 1459 bases. It is submitted that this could be carried out by the skilled person on the basis of the disclosure of D2 in combination with the teaching of D1 to find the essential sequences of the full functional promoter . An inventive step is therefore denied for the subject-matter of all claims of the examined invention (Article 33(3) PCT).

5. None of the present claims of the examined invention is limited by the applied wording to the promoter sequence as present in hProm505, demonstrated in Fig. 5A to be a particularly effective promoter. A claim clearly limited to the promoter sequence of 505 nucleotides in the hTR gene may be considered to involve an inventive step (present claims 1 and 2 are not excluding the presence of additional 5' end sequences of the hTR gene in the isolated promoter sequence by the use of the wording "having" in stead of "consisting": see page 5, last paragraph).

It is noted that it is an error is made in the last paragraph on page 5 as reference

is made to position -463 as shown in Fig.4(A); this should be position -436.

Re Item VI

Certain documents cited

Certain published documents (Rule 70.10)

Application No / Patent No	Publication date (day/month/year)	Filing date (day/month/year)	Priority date (valid claim) (day/month/year)
WO 98 11207 (D5)	19.03.98	16.09.97	16.09.96

Re Item VIII

Certain observations on the international application

6. In the present report the documents D4-D5 cited in the International Search Report as "P,X-document" have not been considered as the priority document is not available.

****

Claims

1. An isolated promoter sequence derived from the telomerase RNA (TR) gene promoter, having approximately 505 bp upstream of the transcription start site or a fragment thereof, capable of initiating transcription of DNA operably linked downstream of said promoter.

2. An isolated promoter sequence according to claim 1 wherein the promoter sequence is construct hProm505 as shown in Fig 4a and Fig 5a.

3. An isolated promoter sequence according to claim 1 or claim 2 wherein the promoter sequence is 230 bp in length starting at position -42 bp as shown in Fig 4a and Fig 5a upstream of the transcription start site.

4. An isolated promoter sequence according to any one of the preceding claims having the sequence as shown in Fig 4a or mutant, allele, derivative or variant thereof.

5. An isolated promoter sequence according to any one of the preceding claims operably linked to a heterologous nucleic acid coding sequence or a gene.

6. A nucleic acid construct comprising a promoter sequence according to any one of claims 1 to 4, operably linked to a heterologous gene.

7. A nucleic acid construct according to claim 6 wherein the heterologous gene encodes a cytotoxin.

8. A vector comprising an isolated promoter sequence according to any one of claims 1 to 5 or a nucleic acid construct according to claim 6 or claim 7.

9. A host cell comprising an isolated promoter sequence according to any one of claims 1 to 5 or a vector according to claim 8.

10. A host cell comprising a nucleic acid construct according to claim 6 or claim 7.

11. A method comprising culturing a host cell according to claim 10 under conditions for transcription of said heterologous sequence from the promoter.

12. A method according to claim 11 wherein the heterologous sequence is a coding sequence and the host cell is cultured under conditions for expression of the encoded peptide or polypeptide product.

13. A method according to claim 10 or claim 11 comprising detection of transcription of the heterologous sequence or the encoded product.

14. A method of screening for ability of a substance to modulate activity of a TR promoter, the method comprising contacting an expression system containing a nucleic acid construct according to claim 6 with a test or candidate substance and determining transcription of the heterologous sequence or expression of the encoded peptide or polypeptide product.

15. A method according to claim 14 wherein the expression system comprises a host cell containing said nucleic acid construct.

16. A method which comprises, following identification of a substance having the ability to modulate activity of a TR promoter in accordance with a method according to claim 14 or claim 15, manufacture of the substance and/or use of the substance in manufacture or formulation of a composition.

17. A vector according to claim 8 for use in a method of gene therapy.

18. Use of a vector according to claim 8 in the preparation of a medicament for the treatment of cancer.

19. A system for use in the control of neoplasia in a human or animal subject comprising a vector or other

delivery system capable of selectively infecting tumour cells in said subject, said vector carrying a DNA or RNA sequence encoding an enzyme operably linked to a nucleic acid molecule according to any one of claims 1 to 5, in association with a prodrug capable of being converted to an active compound by action of said enzyme.

20. A system according to claim 19 wherein the vector is the gene therapy vector pGT62-codAupp.

21. A system according to claim 19 or 20 wherein the enzyme is viral thymidine kinase.

22. A system according to claim 19 or 21 wherein the prodrug is ganciclovir.

23. Use of a system according to any one of claims 19 to 22 in the preparation of a medicament for the treatment of neoplasia in a human or animal subject.

24. A method of treating neoplasia in a human or animal subject requiring such treatment comprising administering to the subject an effective amount of a prodrug and a modified virus capable of selectively infecting tumour cells in said subject, said virus carrying a DNA or RNA sequence encoding an enzyme capable of converting said produg to an active compound, wherein said DNA or RNA

sequence is operably linked to a nucleic acid molecule according to any one of claim 1 to 5.

25. A method according to claim 24 wherein the enzyme is viral thymidine kinase and the prodrug is ganciclovir.

REPLACED BY ART 34 AMDT

Claims

1. A nucleic acid molecule comprising a promoter sequence derived from the telomerase RNA (TR) gene promoter.

2. A nucleic acid molecule according to claim 1 wherein the promoter sequence is derived from the human TR (hTR) gene promoter.

3. A nucleic acid molecule according to claim 1 or claim 2 wherein the promoter sequence is derived from a nucleotide sequence as shown in Fig. 4a.

4. A nucleic acid molecule according to any one of the preceding claims wherein the promoter sequence is at least 230bp in length starting at position -42bp as shown in Fig. 4a and Fig 5a upstream of the transcription start site.

5. A nucleic acid molecule according to any of the preceding claims wherein the promoter sequence comprises the construct hProm505 as shown in Fig.4a and Fig.5a.

6. A nucleic acid molecule according to any one of the preceding claims wherein the promoter sequence comprises the construct hProm867 as shown in Fig. 4a and Fig. 5a.

7. A nucleic acid molecule according to any one of the preceding claims wherein the promoter sequence comprises the nucleotide sequence as shown in Fig. 4a.

8. A nucleic acid molecule according to claim 1 wherein the promoter sequence is derived from mouse TR (terc) gene promoter.

9. A nucleic acid molecule according to claim 8 wherein the promoter sequence is derived from the nucleotide sequence as shown in Fig. 4b.

10. A nucleic acid molecule according to claim 8 or claim 9 wherein the promoter sequence is at least 73bp in length starting at -22bp as shown in Fig. 4b and Fig. 5a upstream of the transcription start site.

11. A nucleic acid molecule according to any one of claims 8 to 10 wherein the promoter sequence comprises the construct mProm208 as shown in Fig. 4b and Fig. 5b.

12. A nucleic acid molecule according to any one of claims 8 to 11 wherein the promoter sequence comprises the construct mProm628 as shown in Fig. 4b and Fig. 5b.

13. A nucleic acid molecule according to any one of claims 8 to 12 wherein the promoter sequence comprises a nucleotide sequence as shown in Fig. 4b.

14. A nucleic acid molecule having promoter activity and being capable of hybridising under stringent conditions to the complementary sequence of a nucleic acid molecule according to any one of the preceding claims.

15. A nucleic acid construct comprising a TR promoter

region or a fragment, mutant, allele derivative or variant thereof able to promote transcription, operably linked to a heterologous gene.

16. A nucleic acid construct according to claim 15 wherein the heterologous gene encodes a cytotoxin.

17. A vector comprising a nucleic acid molecule according to any one of claims 1 to 14 or a nucleic acid construct according to claim 15 or claim 16.

18. A host cell comprising a nucleic acid molecule according to any one of claims 1 to 14 or a vector according to claim 17.

19. A host cell comprising a nucleic acid construct according to claim 15 or claim 16.

20. A method comprising culturing a host cell according to claim 19 under conditions for transcription of said heterologous sequence from the promoter.

21. A method according to claim 20 wherein the heterologous sequence is a coding sequence and the host cell is cultured under conditions for expression of the encoded peptide or polypeptide product.

22. A method according to claim 20 or claim 21 comprising detection of transcription of the heterologous sequence or the encoded product.

23. A method of screening for ability of a substance to modulate activity of a TR promoter, the method comprising contacting an expression system containing a nucleic acid construct according to claim 15 with a test or candidate substance and determining transcription of the heterologous sequence or expression of the encoded peptide or polypeptide product.

24. A method according to claim 23 wherein the expression system comprises a host cell containing said nucleic acid construct.

25. A method which comprises, following identification of a substance a having the ability to modulate activity of a TR promoter in accordance with a method according to claim 23 or claim 24, manufacture of the substance and/or use of the substance in manufacture or formulation of a composition.

26. A substance having the ability to modulate activity of a TR promoter as determined by telomerase RNA gene expression.

27. A substance according to claim 26 which specifically blocks transcriptional activation of the TR gene promoter through interaction of the 5' regulatory sequences.

28. A substance according to claim 26 or claim 27 being selected from the group consisting of antisense oligonucleotides, transcription factors, peptide nucleic acids, and factors that disrupt signal transduction

pathways.

29. A substance according to any one of claims 26 to 28 which is Sp3 transcription factor or the CCAAT protein complex.

30. A substance according to any one of claims 26 to 29 for use in medical treatment.

31. Use of a substance according to any one of claims 26 to 29 in the preparation of a medicament for the treatment of cancer.

32. A substance according to claim 26 which specifically activates the TR gene promoter through interaction with the 5' regulatory sequences.

33. A substance according to claim 32 which is Sp1 transcription factor or pRb transcription factor.

34. Use of a substance according to claims 32 or 33 for the preparation of a medicament for the treatment of cancer.

35. Use of a substance according to any one of claims 26 to 29 in the preparation of a medicament for repressing the TR gene promoter in cells.

36. Use of a substance according to claims 32 or 33 for the preparation of a medicament for activating the TR gene promoter in cells.

37. A vector according to claim 17 for use in a method of gene therapy.

38. Use of a vector according to claim 17 in the preparation of a medicament for the treatment of cancer.

39. A system for use in the control of neoplasia in a human or animal subject comprising a vector or other delivery system capable of selectively infecting tumour cells in said subject, said vector carrying a DNA or RNA sequence encoding an enzyme operably linked to a nucleic acid molecule according to any one of claims 1 to 14, in association with a prodrug capable of being converted to an active compound by action of said enzyme.

40. A system according to claim 39 wherein the vector is the gene therapy vector pGT62-codAupp.

41. A system according to claim 39 or 40 wherein the enzyme is viral thymidine kinase.

42. A system according to claim 39 or 41 wherein the prodrug is ganciclovir.

43. Use of a system according to any one of claims 39 to 42 in the preparation of a medicament for the treatment of neoplasia in a human or animal subject.

44. A method of treating neoplasia in a human or animal subject requiring such treatment comprising administering to the subject an effective amount of a prodrug and a

modified virus capable of selectively infecting tumour cells in said subject, said virus carrying a DNA or RNA sequence encoding an enzyme capable of converting said produg to an active compound, wherein said DNA or RNA sequence is operably linked to a nucleic acid molecule according to any one of claim 1 to 14.

45. A method according to claim 44 wherein the enzyme is viral thymidine kinase and the prodrug is ganciclovir.

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International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 6 :
C12N 15/11, 9/12, 15/85, C12Q 1/68

A2

(11) International Publication Number: WO 99/38964

(43) International Publication Date: 5 August 1999 (05.08.99)

(21) International Application Number: PCT/GB99/00308

(22) International Filing Date: 29 January 1999 (29.01.99)

(30) Priority Data:
9801902.9 29 January 1998 (29.01.98) GB

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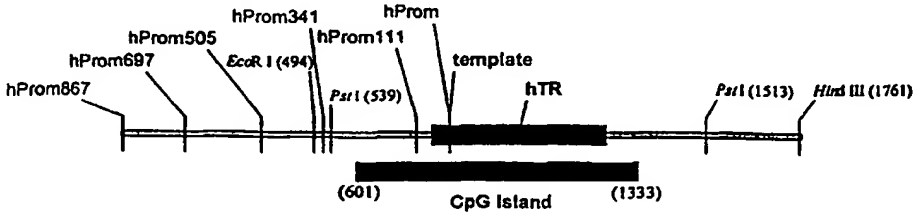
(81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published
Without international search report and to be republished upon receipt of that report.

(54) Title: PROMOTER REGIONS OF THE MOUSE AND HUMAN TELOMERASE RNA COMPONENT GENES

(57) Abstract

The present invention relates to the identification of the genomic promoter region of the human and mouse telomerase RNA gene. Telomerase activity is necessary for the unrestricted proliferative capacity of many human cancers. It is proposed that mutation or dysregulation of the telomerase repression pathway may cause reactivation or upregulation of telomerase expression in cancer. The invention provides details of elements important for the regulation of telomerase RNA genes, including the Sp family of transcription factors. There is further provided methods for screening for elements having the ability for suppressing telomerase RNA gene promoter activity and use of such elements in the treatment of cancers. In addition, evidence is also provided for the development of new transcription based therapies for cancer and for genetic approaches to targeting therapeutic genes to cancer cells. Namely, (1) transcriptional repression and the disruption of signal transduction pathways regulating telomerase activation. (2) Tumour specific gene expression for genetic therapy via telomerase RNA gene promoters.



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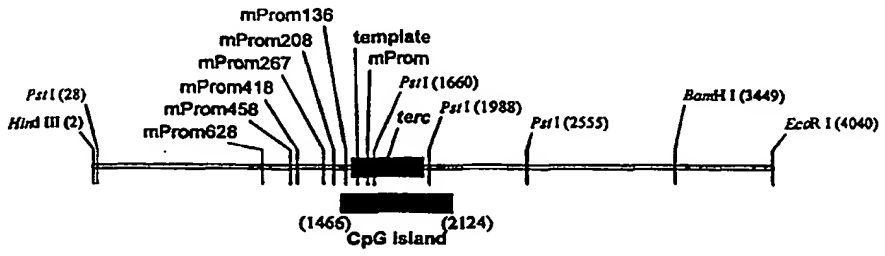

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Fig. 1

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Fig. 2

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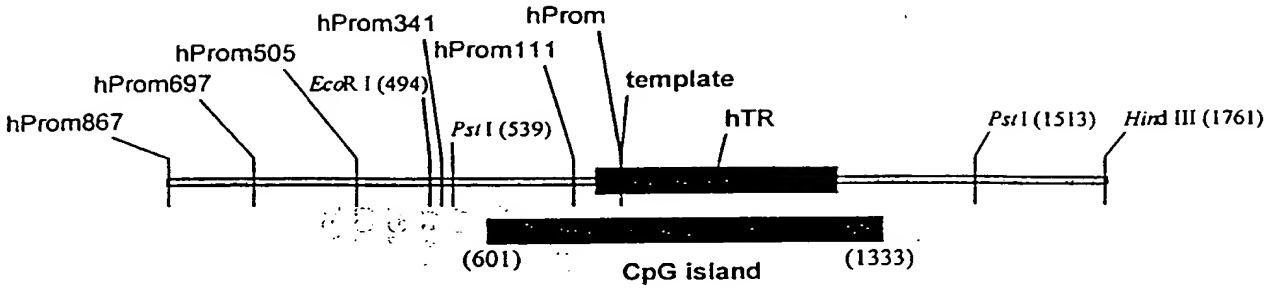
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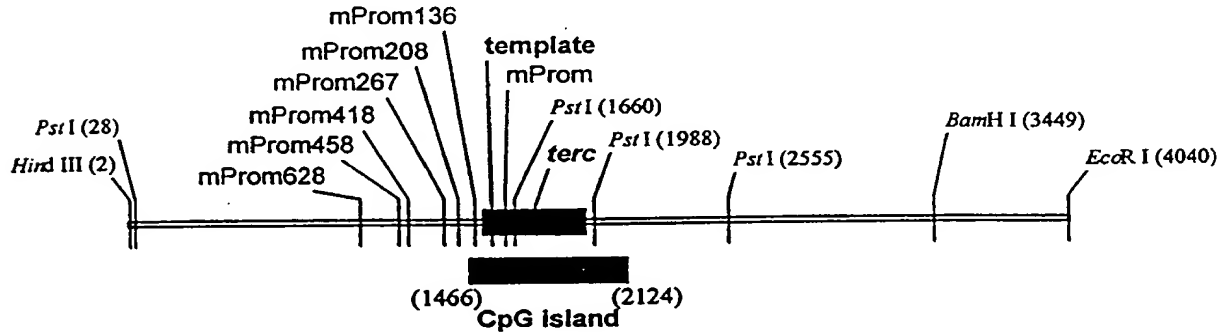
Fig. 3A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(1466)
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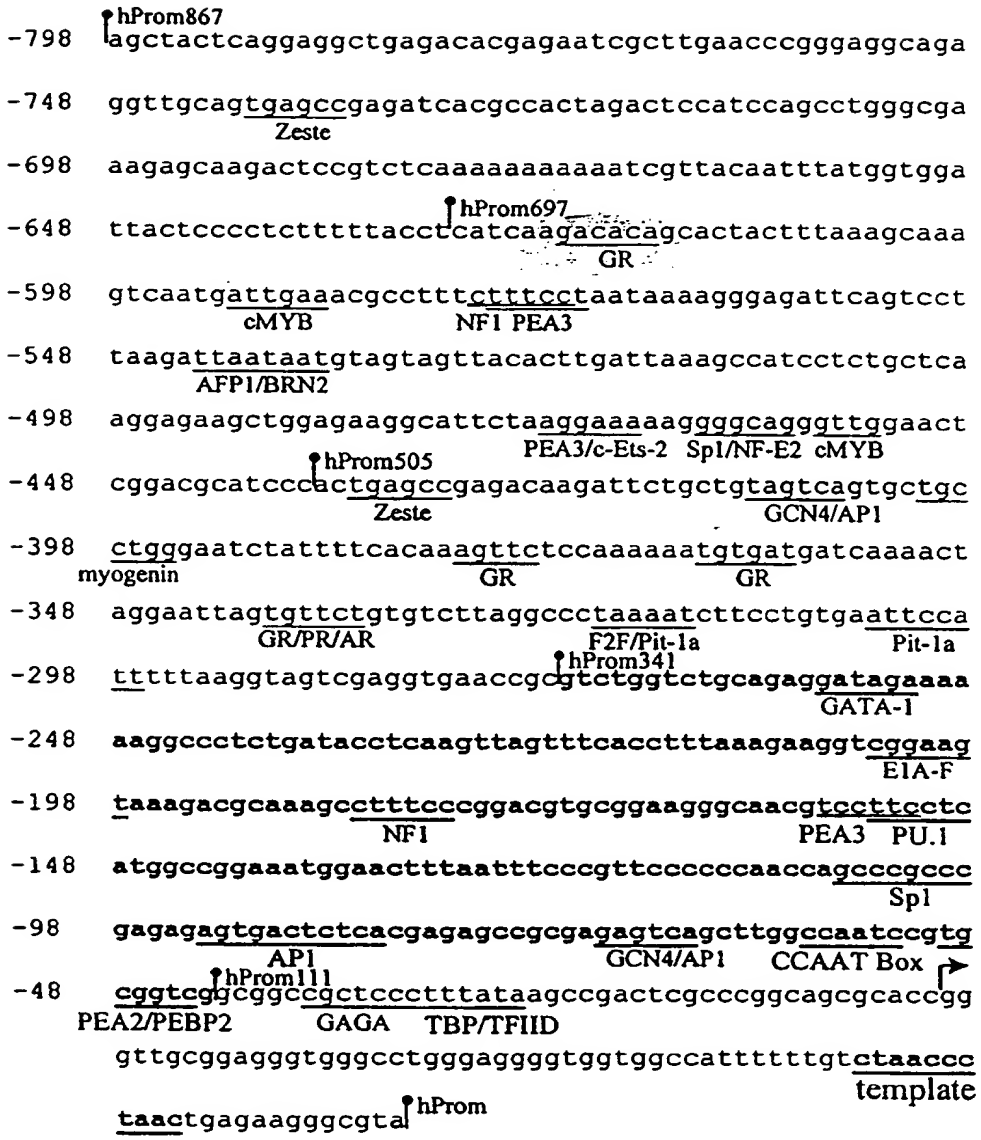
CpG island

Fig. 3B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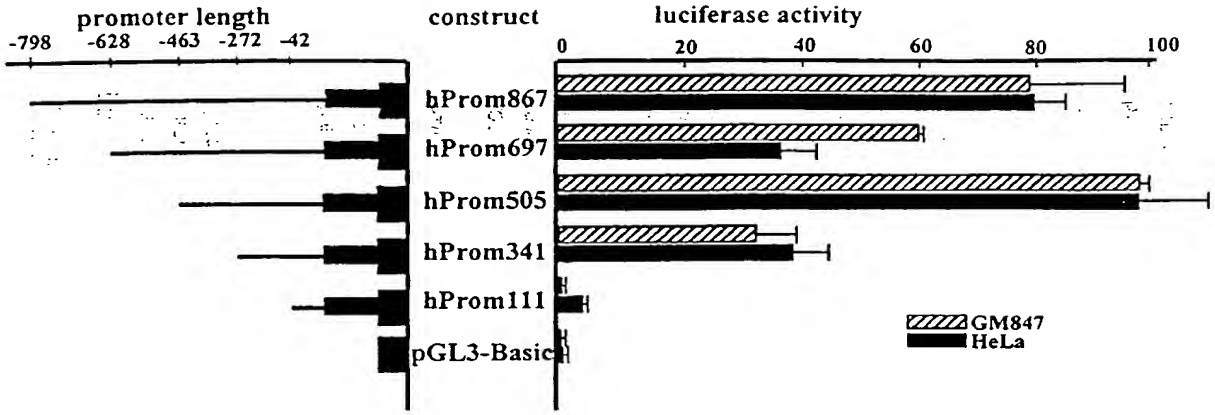
-748 ggttgcagtgagccgagatcacgccactagactccatccagcctgggcga
Zeste
-698 aagagcaagactccgtctcaaaaaaaaaaatcgttacaatttatggtgga
hProm697
-648 ttactcccctcttttacctcatcaagacacagcactactttaaagcaaa
GR
-598 gtcaatgattgaaacgcctttctttcctaataaaagggagattcagtcct
cMYB
NF1 PEA3
-548 taagattaataatgtagtagttacacttgattaaagccatcctctgctca
AFP1/BRN2
-498 aggagaagctggagaaggcattctaaggaaaaaggggcagggttggaact
PEA3/c-Ets-2 Sp1/NF-E2 cMYB
hProm505
-448 cggacgcatcccactgagccgagacaagattctgctgtagtcagtgctgc
Zeste
GCN4/AP1
-398 ctgggaatctattttcacaaagttctccaaaaaatgtgatgatcaaaact
myogenin
GR
GR
-348 aggaattagtgttctgtgtcttaggccctaaaatcttcctgtgaattcca
GR/PR/AR
F2F/Pit-1a
Pit-1a
hProm341
-298 tttttaaggtagtcgaggtgaaccgcgtctggtctgcagaggatagaaaa
GATA-1
-248 aaggccctctgatacctcaagttagtttcacctttaaagaaggtcggaag
E1A-F
-198 taaagacgcaaagcctttcccggacgtgcggaagggcaacgtccttcctc
NF1
PEA3 PU.1
-148 atggccggaaatggaactttaatttcccgttcccccaaccagcccgccc
Sp1
-98 gagagagtgactctcacgagagccgcgagagtcagcttggccaatccgtg
AP1
GCN4/AP1
CCAAT Box
hProm111
-48 cggtcggcggccgctccctttataagccgactcgcccggcagcgcaccgg
PEA2/PEBP2
GAGA
TBP/TFIID
gttgcggagggtgggcctgggaggggtggtggccatttttgtctaaccc
template
taactgagaagggcgta
hProm

Fig. 4A


mProm628
-531 tgtgaccttgaactacagacctcctgcctcagcctcctacaagctgggat
PPAR/ELP GR
-481 tataggctcgggtcagctacccttgaaatctttttctttctggaactcag
H4TF-1
-431 tacctggttggccatgcactcacaagagatccgcctgccttctgtctctc
AP1/GCN4/Zeste SP-1
mProm458
-381 aaattctggaattaaagatgtgcgccactttccccacttccacccccgg
C/EBPalpha, beta AP-2
mProm418
-331 ctgtgggagtggactgggttgaaggtggaatttttttttttttttttt
p300
-281 ttttagtgaaaaaaggggggattggaaatatccctactttcaactctagt
CP1
-231 atatttcagaaaccaagcctcagagatgtgcgtgcgtgcgtgtgtgtgtg
mProm267
-181 tgtatgtgtgtgtgtgtctcacagcaagaaacagattttattatttatttttt
GR F2F/Pit-1a
mProm208
-131 tatttatttatttttgcaagtgactggctaggaagagtggggaagcggg
IRF-1,2 GCN4/AP1 AP-2 c-ETS-2
mProm136
-81 aggacaaatggggaagagggagcatttccgcaagtgctgggctcgaccaa
GR AP-2 c-ETS-2 AP-2 CCAAT Box/AP-1
-31 tcagcgcgcgccatggggtatttaaggtcgagggcggctaggcctcggca
cctaaccctgattttcattagctgtgggttctggtcttttgttctccgcc
template
cgctgtttttctcgctgacttccagcgg
mProm

Fig. 4B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construct
luciferase activity
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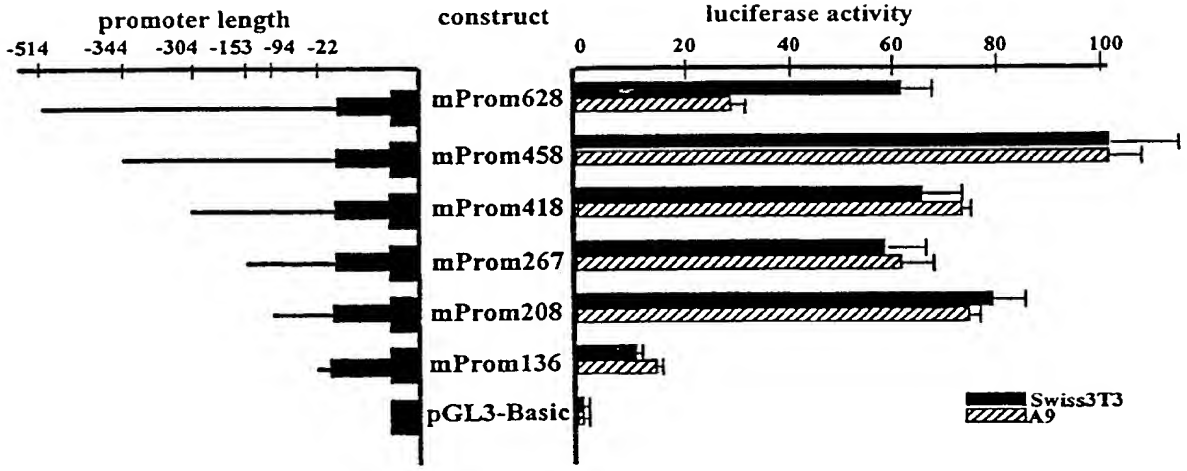
hProm867
hProm697
hProm505
hProm341
hProm111
pGL3-Basic
GM847
HeLa

Fig. 5A


promoter length
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construct
luciferase activity
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mProm458
mProm418
mProm267
mProm208
mProm136
pGL3-Basic
Swiss3T3
A9

Fig. 5B

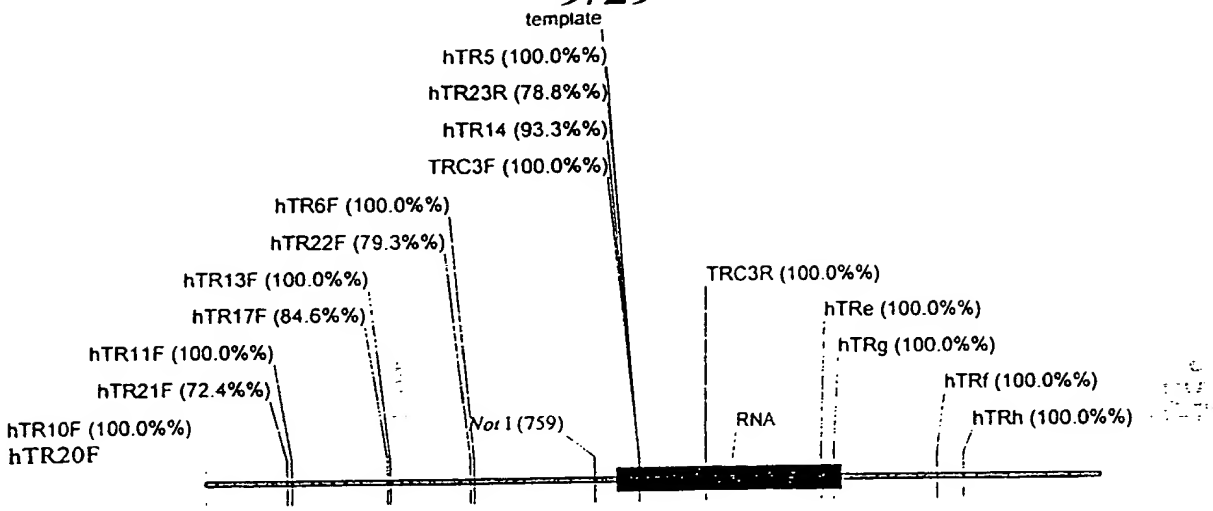
Oligo's Used: human

Name	Sequence	Comments
hTR5	TACGCCCTTCTCAGTTAGGGTTAG	
hTR14	GGATCCTACGCCCTTCTCAGTTAGGGTTAG	hTR5 with BamHI site
hTR13F	ACTGAGCCGAGACAAGATTC	
hTR17F	GGATCCACTGAGCCGAGACAAGATTC	hTR13F with BamHI site
hTR10F	AGCTACTCAGGAGGCTGAGA	
hTR20F	GCGCTCGAGAGCTACTCAGGAGGCTGAGA	hTR10F with XhoI site plus gcg clamp
hTR11F	CATCAAGACACAGCACTACT	
hTR21F	GCGCTCGAGCATCAAGACACAGCACTACT	hTR11F with XhoI site plus gcg clamp
hTR6F	GTCTGGTCTGCAGAGGATAG	
hTR22F	GCGCTCGAGGTCTGGTCTGCAGAGGATAG	hTR6F with Xho site plus gcg clamp
hTR5	TACGCCCTTCTCAGTTAGGGTTAG	
hTR23R	CGCAAGCTTTACGCCCTTCTCAGTTAGGGTTAG	hTR5 with HindIII site plus cgc clamp
hTRe	CTGAGCTGTGGGACGTGCAC	
hTRf	AGACGGGAGAACCCACGCAG	
hTRg	CTCGGCTCACACATGCAGTT	
hTRh	TCTGCAGAGCAGGAACTAAGT	
TRC3F	CTAACCCTAACTGAGAAGGGCGTA	
TRC3R	GGCGAACGGGCCAGCAGCTGACATT	

Oligo's Used: Mouse

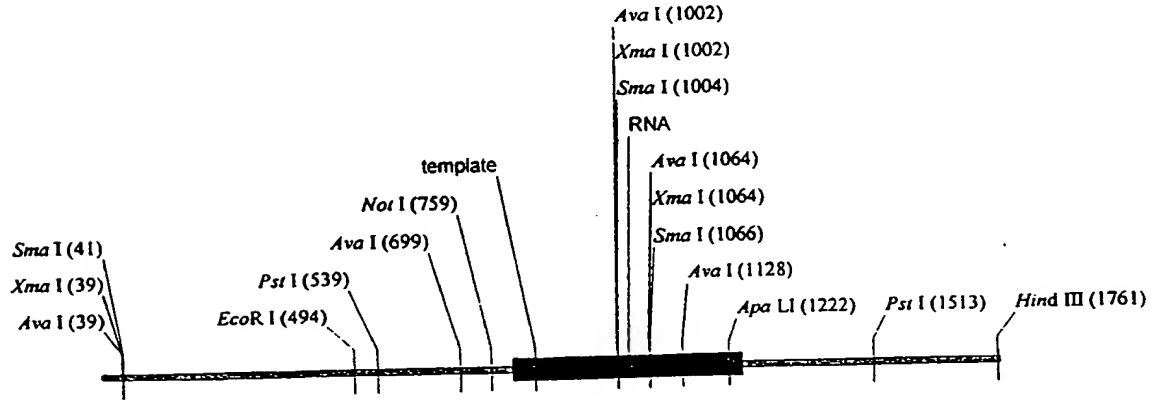
Name	Sequence	Comments
mTR16F	GTGTCTCACAGCAAGAAACA	
mtr25f	GCGCTCGAGGTGTCTCACAGCAAGAAACA	This is mtr16f with XhoI site plus gcg clamp
mTR17F	GTGACTGGCTAGGAAGAGTG	
mtr26f	GCGCTCGAGGTGACTGGCTAGGAAGAGTG	This is mtr17f with XhoI site plus gcg clamp
mTR18F	TGTGACCTTGAACTACAGAC	
mtr27f	GCGCTCGAGTGTGACCTTGAACTACAGAC	This is mtr18f with XhoI site plus gcg clamp
mTR19F	GGACTGGGTTGAAGGTGGAA	
mtr28f	GCGCTCGAGGGACTGGGTTGAAGGTGGAA	This is mtr19f with XhoI site plus gcg clamp
mTR20F	TGCGCCACTTTTCCCCACTT	
mtr29f	GCGCTCGAGTGCGCCACTTTTCCCCACTT	This is mtr20f with XhoI site plus gcg clamp
mTRr1	CCGCTGGAAGTCAGCGAGAA	
mtr30r	CGCAAGCTTCCGCTGGAAGTCAGCGAGAA	This is mTRr1 with HindIII site plus cgc clamp
mTR36F	GCGCTCGAGTCGACCAATCAGCGCGCGCCAT	This is Xho I site PCR primer plus gcg clamp
mTRr1	CCGCTGGAAGTCAGCGAGAA	
mTRf1	TCGACCAATCAGCGCGCGCCAT	

Fig. 6



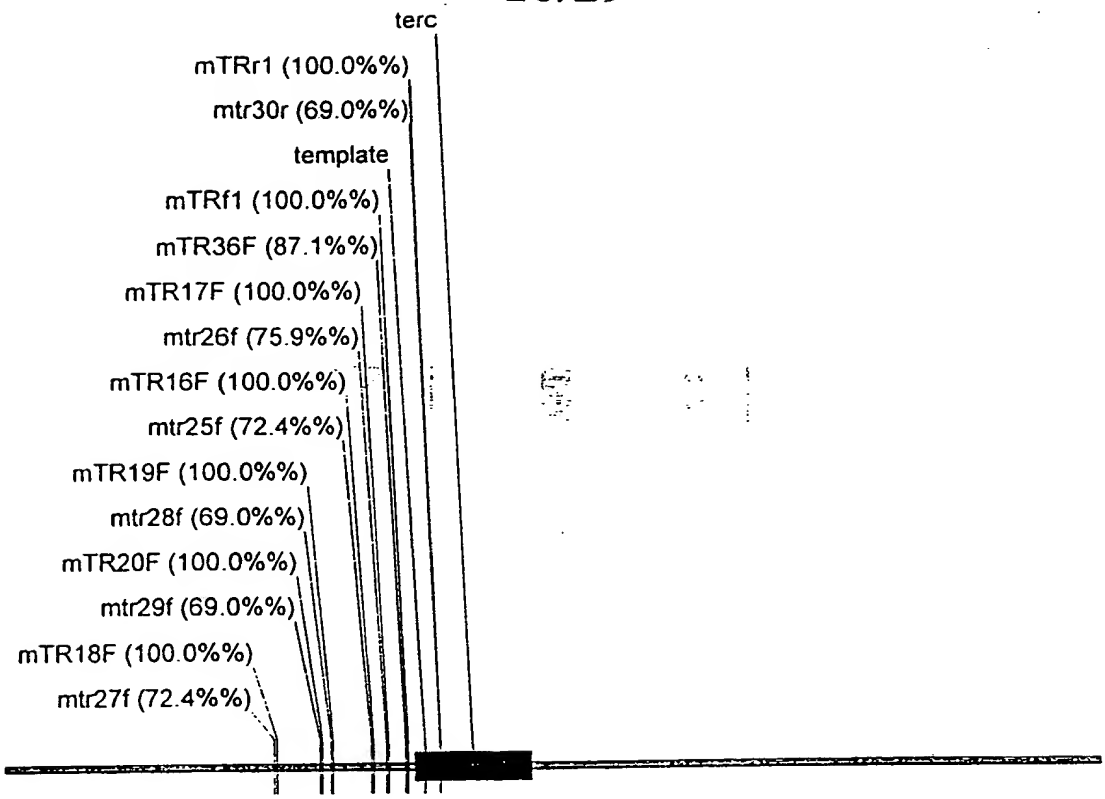
htrcon
1765 bp

Fig. 7A



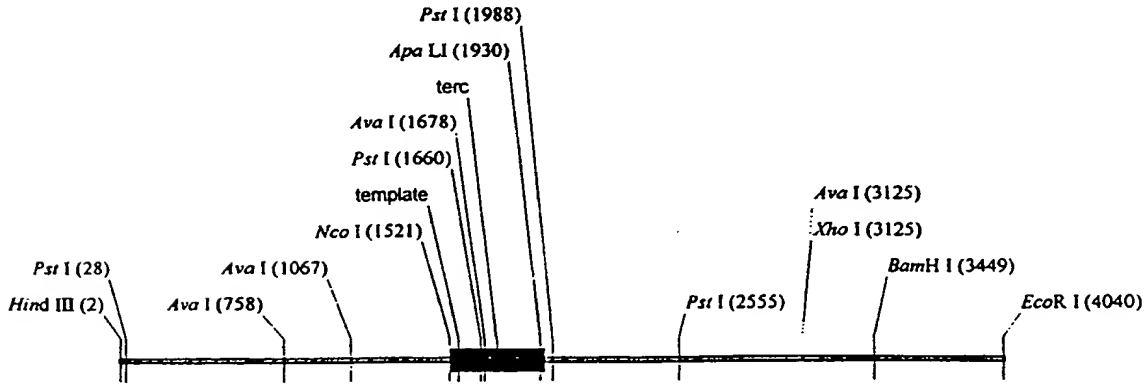
htrcon
1765 bp

Fig. 7B



tercCon
4044 bp

Fig. 8A

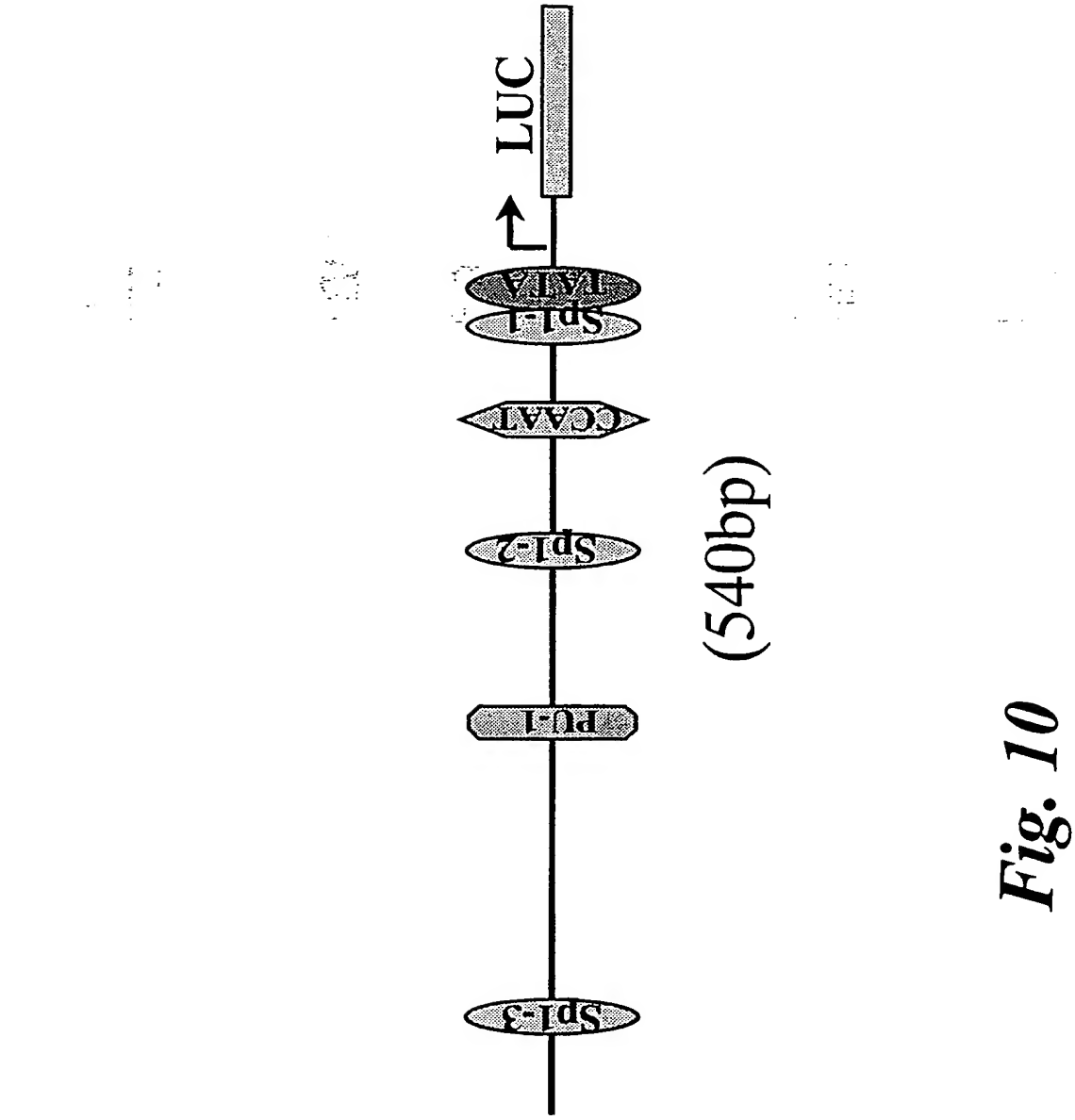


tercCon
4044 bp

Fig. 8B

TTGTGACCTTGAACTACAGACCTCCTGCCTCAGCCTCCTACAAGCTGGGATTATAGGCTCGGGTCAGCTACCCTTGAAA
TTCTTTTTCTTTCTGGAACTCAGTACCTGGTTGGCCATGCACTCACAAGAGATCCGCCTGCCTTCTGTCAAATTCTGGA
ATTAAAGATTTGCGCCACTTTTCCCCACTTCCACCCCCGGCTGTGGGAGTGGACTGGGTTGAAGGTGGAATTTTTTTTT
TTTTTTTTTTAGTGAAAAAAGGGGGGATTGGAAATATCCCTACTTTCAACTCTAGTATATTTCAGAAACCAAGCCTCAG
AAATGTGCGTGCGTGCGTGTGTGTGTGTGTATGTGTGTGTGTCTCACAGCAAGAAACAGATTTTATTATTTATTTTTA
TTTATTTATTTTTTGCAAGTGACTGGCTAGGAAGAGTGGGGAAGCGGGAGGACAAATGGGGAAGAGGGAGCATTTCCGC
AAGTGCTGGGCTCGACCAATCAGCGCGCGCCATGGGGTATTTAAGGTCGAGGGCGGCTAGGCCTCGGCACCTAACCCTG
ATTTTCATTAGCTGTGGGTTCTGGTCTTTCGTTCTCCGCCCGCTGTTTTTCTCGCTGACTTCCAGCGGA

Fig. 9



Human Promoter
TATA
CCAAT
SP-1
AP1
PEA2/PEBP2
PU.1
PEA3
GATA1
C-MYB
GR/PR/AR
F2F
ZESTE

Fig. 10

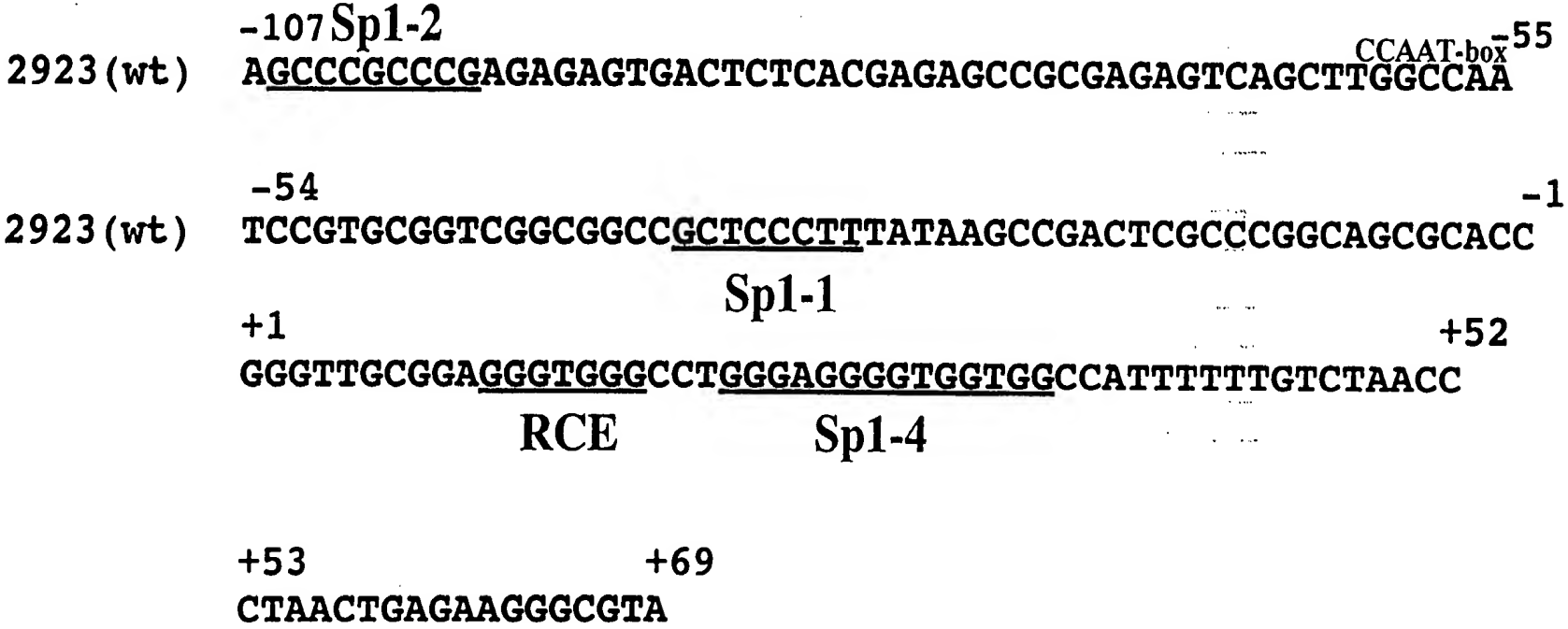

-107 Sp1-2
CCAAT-box -55
2923(wt) AGCCCGCCCGAGAGAGTGACTCTCACGAGAGCCGCGAGAGTCAGCTTGGCCAA
-54 -1
2923(wt) TCCGTGCGGTCGGCGGCCGCTCCCTTTATAAGCCGACTCGCCCGGCAGCGCACC
Sp1-1
+1 +52
GGGTTGCGGAGGGTGGGCCTGGGAGGGGTGGTGGCCATTTTTTGTCTAACC
RCE Sp1-4
+53 +69
CTAACTGAGAAGGGCGTA

Fig. 11

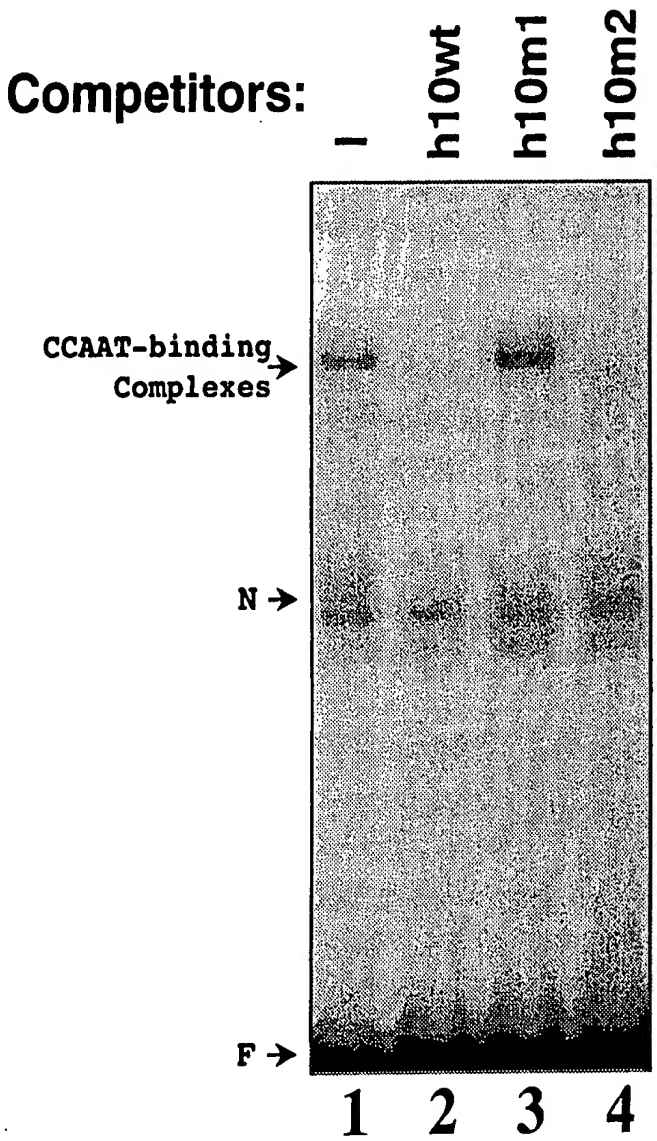
Oligonucleotide	Position	Sequence*	Purpose
h11[a]	-2 to +36	CCGGGTTGCGGAGGGTGGGCCTGGGAGGGGTGGTGGCC	RCE(+12,+16 and +30, +34) and Sp1.4 binding site
h111[a]	"	CCGGGTTGCGGAAAATGGGCCTGGGAGGGGTGGTGGCC	RCE1 mutation from ggg to aaa (+11/+13,)
h112[a]	"	CCGGGTTGCGGAGGGTGGGCCTGGGTAAGGTGGTGGCC	Sp1.4 mutation from agg to taa (+24/+26)
h113[a]	"	CCGGGTTGCGGAAAATGGGCCTGGGTAAGGTGGTGGCC	Mutant of both RCE1 and Sp1.4 binding site(+11/+13, +24/+26)
h11c[a]	-2 to +23	CCGGGTTGCGGAGGGTGGGCCTGGG	RCE1 binding site
H11d[a]	+14 to +36	GCCTGGGAGGGGTGGTGGCC	Sp1.4 or RCE2 binding site
h111a[a,b]	"	CCGGGTTGCGGAAAATGGGCCTGGG	RCE1 mutation from ggg to aaa(+11/+13)
h112b[a,b]	+15 to +36	GGGCCTGGGTAAGGTGGTGGCC	Sp1.4 mutation from agg to taa
h112C[a,b]	"	GGGCCTGGGTAAGGTAATGGCC	Sp1.4 and RCE2 mutant from aggggtgg to taaggtaa(+24/+26, +30/+31)
h11e[a]	"	GGGCCTGGGAGGGGTAATGGCC	RCE2 mutant from gg to (+30/+31)
h10[a]	-63 to -42	CTTGGCCAATCCGTGCGGTCGG	h10 footprinting region containing CCAAT binding site
h101[a]	-63 to -42	CTTGGAGTCTCCGTGCGGTCGG	CCAAT motif mutation from ccaa to agtc, (-58/-55)
h10m11[b]	-74 to-45	GCGAGAGTCAGCTTGGAGTCTCCGTGCGG	CCAAT motif mutation from ccaa to agtc, (-58/-55)
h10m2[a,b]	-63 to -42	CTTGGCCAATCCTGATGGTCGG	h10 mutation from gtgc to tgat, (-51/-47)
h9[a]	-44 to -17	CGGCGGCCGCTCCCTTTATAAGCCGACT	h9 footprinting region containing SP1.1 binding site
h91[a]	-44 to -21	CTTACGCCGCTCCCTTTATAAGCC	h9 mutation from ggcg to ttac, (-43/-40)
h910[b]	-53 to -29	CCGTGCGGTCTTACGCCGCTCCC	h9 mutation from ggcg to ttac, (-43/-40)
h911[a]	-44 to -21	CGGCGTAAACTCCCTTTATAAGCC	h9 mutation from gccg to taaa, (-39/-36)
h92[a]	-44 to -21	CGGCGGCCATAGCCTTTATAAGCC	h9 mutation from gctc to atag, (-36/-33)
h921[a]	-44 to -21	CGGCGGCCGCTCATGCTATAAGCC	h9 mutation from cctt to atgc, (-32/-29)
h93[a]	-44 to -21	CGGCGGCCGCTCCCTTCGACAGCC	h9 mutation from tata to cgac, (-28/-25)
h930[b]	-38 to -14	CCGCTCCCTTCGACAGCCGACTCGC	h9 mutation from tata to cgac, (-28/-25)
h4[a]	-110 to -91	ACCAGCCCGCCCGAGAGAGT	h4 footprinting region containing SP1.2 binding site
h41m[a]	-110 to -91	ACCAGCCCGAACGAGAGAGT	Sp1.2 mutation from cc to aa, (-101/-100)
h5[a]	-471 to –452	GAAAAAGGGGCAGGGTTGGA	SP1.3 binding site
h5m[a]	-471 to -452	GAAAAAGGTTCAGGGTTGGA	Sp1.3 mutation from gg to tt, (-463/-462)

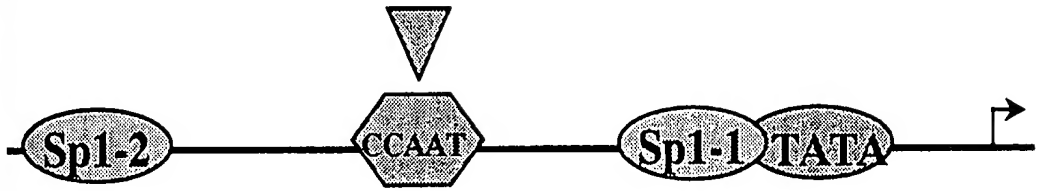
* Nucleotides corresponding to promoter sequences are given in uppercase letters from 5' end to 3' end; specific nucleotides mentioned in Purpose colum are underlined. Highlight indicate mutagenesis nucleotide.

[a] Complementary lowerstrand sequence for EMSA not show.

[b] For PCR-directed in vitro mutagenesis, complementary lowstrand sequence not shown.

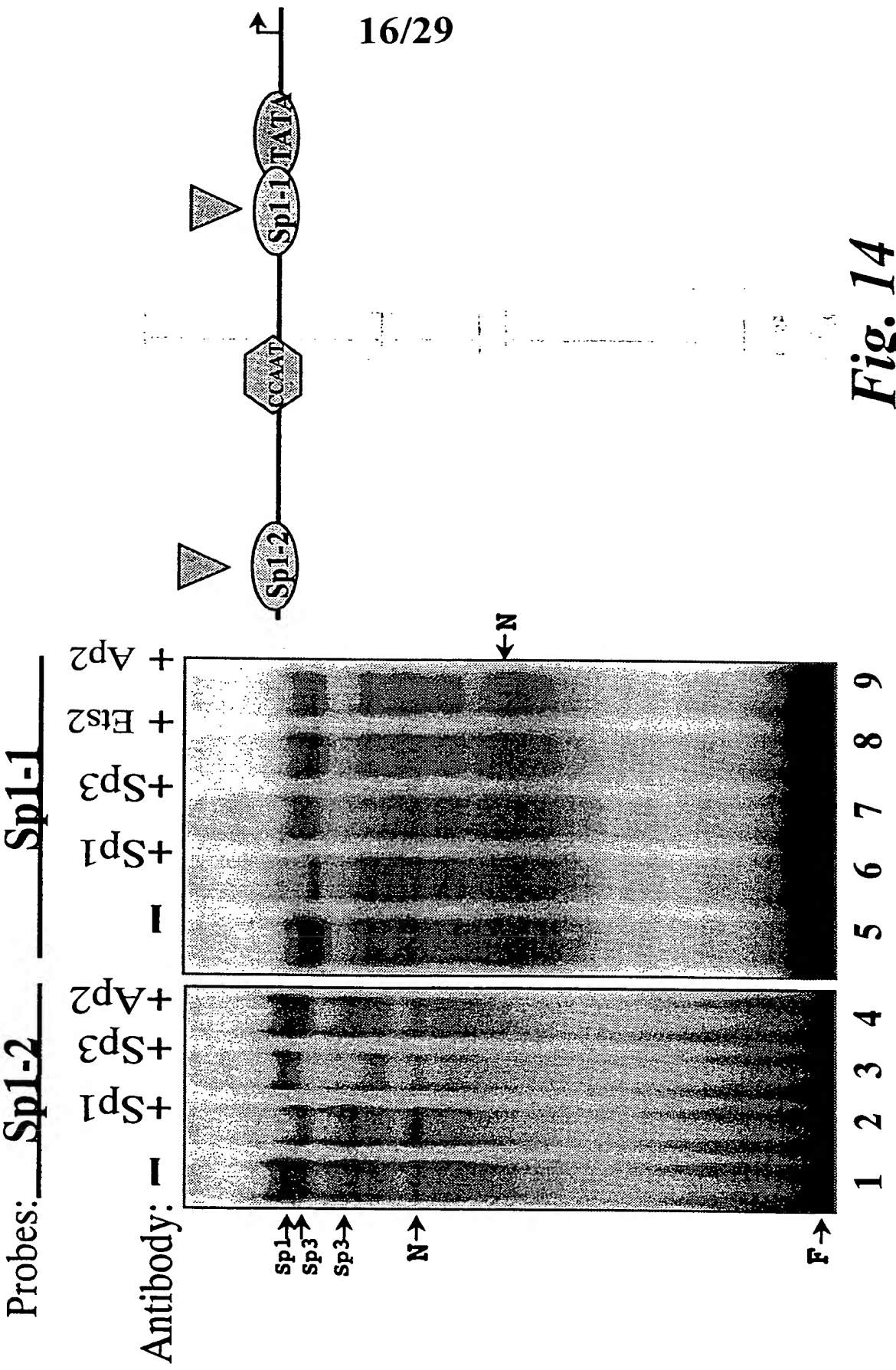
Fig. 12





h10 (wt:-63 -42)	CTTGGCCAATCCGTGCGGTCGG
h10m1 (-58 -55)	:::::AGTC::::::::::::
h10m2 (-51 -47)	::::::::::::TGAT:::::

Fig. 13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TATA
CCAAT
Sp1-2
Probes: Sp1-2 Sp1-1
Antibody: - +Sp1 +Sp3 +Ap2 - +Sp1 +Sp3 +Ets2 +Ap2
N
Sp1
Sp3
Sp3
N
F
1 2 3 4 5 6 7 8 9

Fig. 14

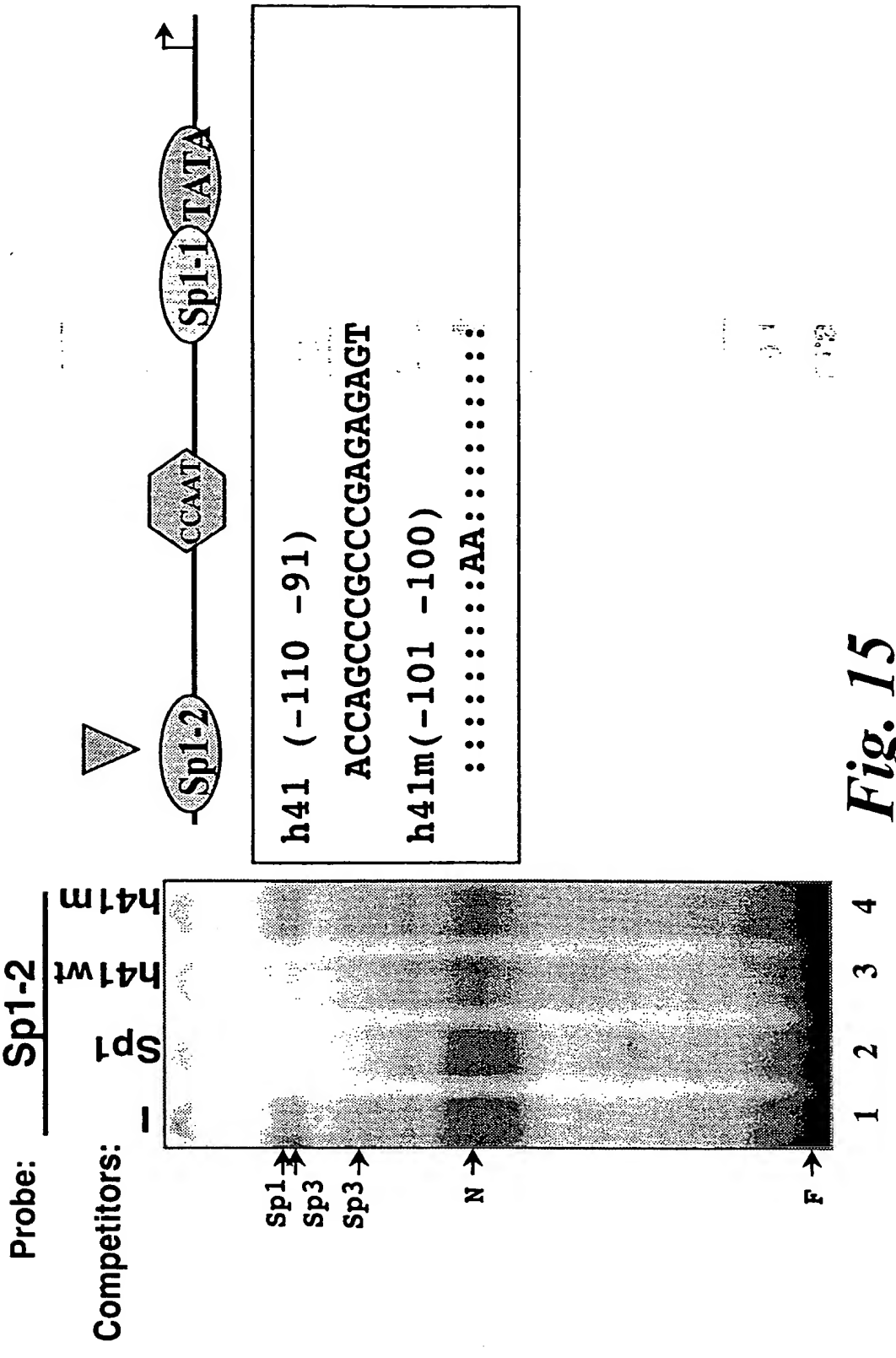
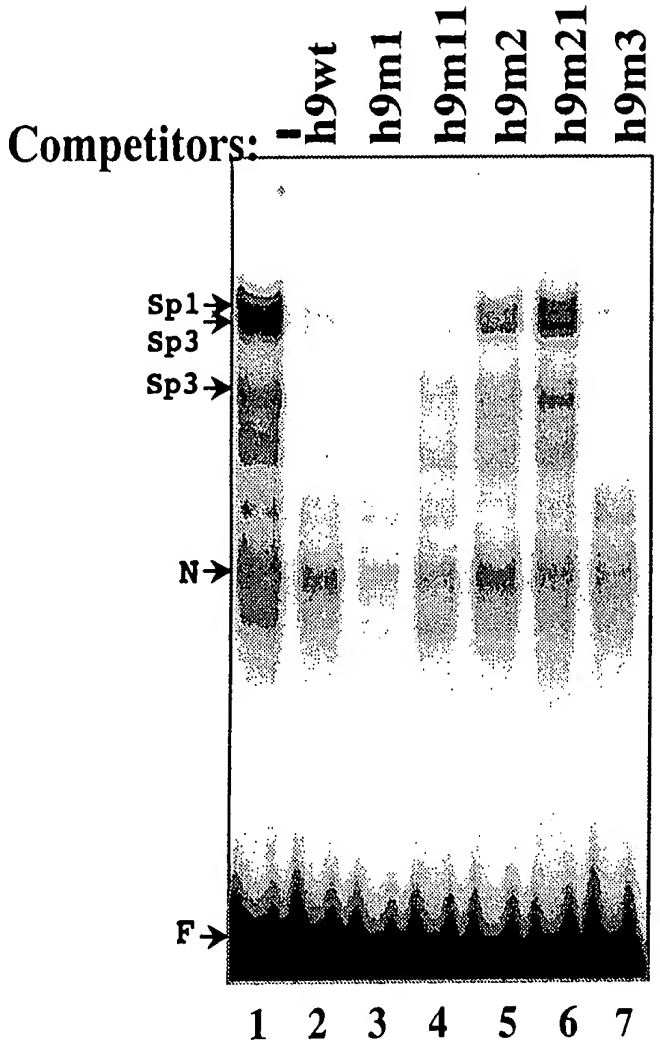
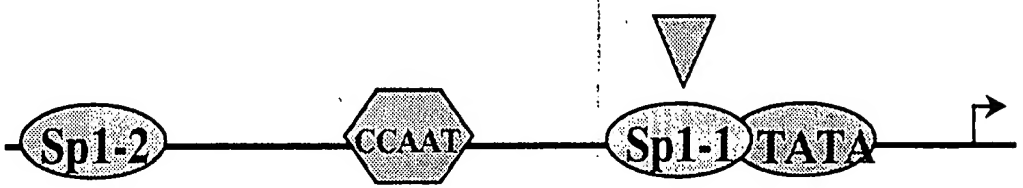

Sp1-2
CCAAT
Sp1-1
TATA
h41 (-110 -91)
ACCAGCCCGCCCGAGAGT
h41m(-101 -100)
::::::::::AA::::::::
Probe:
Sp1-2
Competitors:
-
Sp1
h41wt
h41m
Sp1
Sp3
Sp3
N
F
1 2 3 4

Fig. 15





h9 (-44 -17)	CGGCGGCCGCTCCCTTTATAAGCCGACT
h9m1(-43 -40)	:TTAC::::::::::::::::::::::::
h9m11(-39 -36)	:::::TAAA::::::::::::::::::::
h9m2(-36 -33)	::::::::ATAG::::::::::::::::
h9m21(-32 -29)	::::::::::::ATGC::::::::::::
h9m3(-28 -25)	::::::::::::::::CGAC::::::::

Fig. 16

- Gel shift assays identify DNA/protein binding activity
- Do mutation of these binding sites influence the promoter activity?

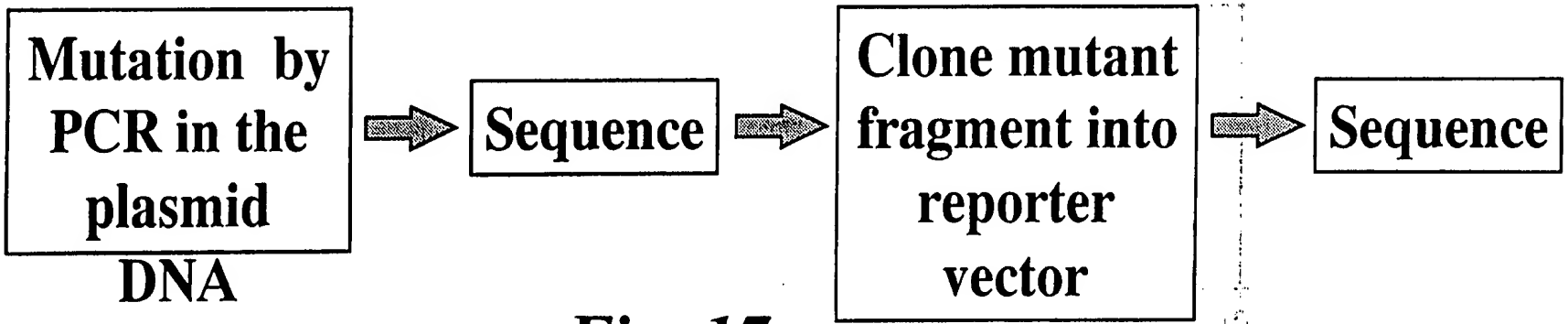


Fig. 17

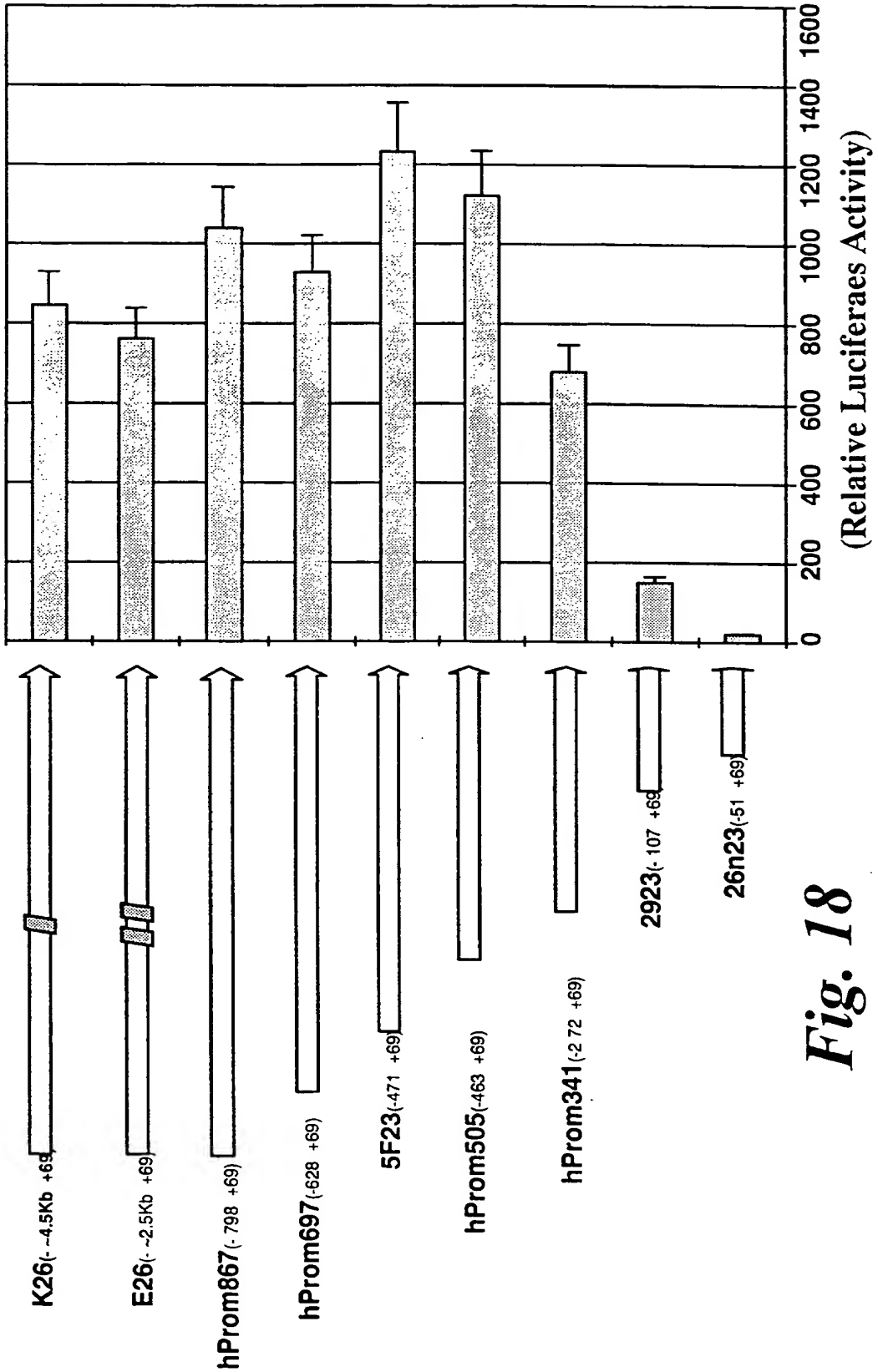
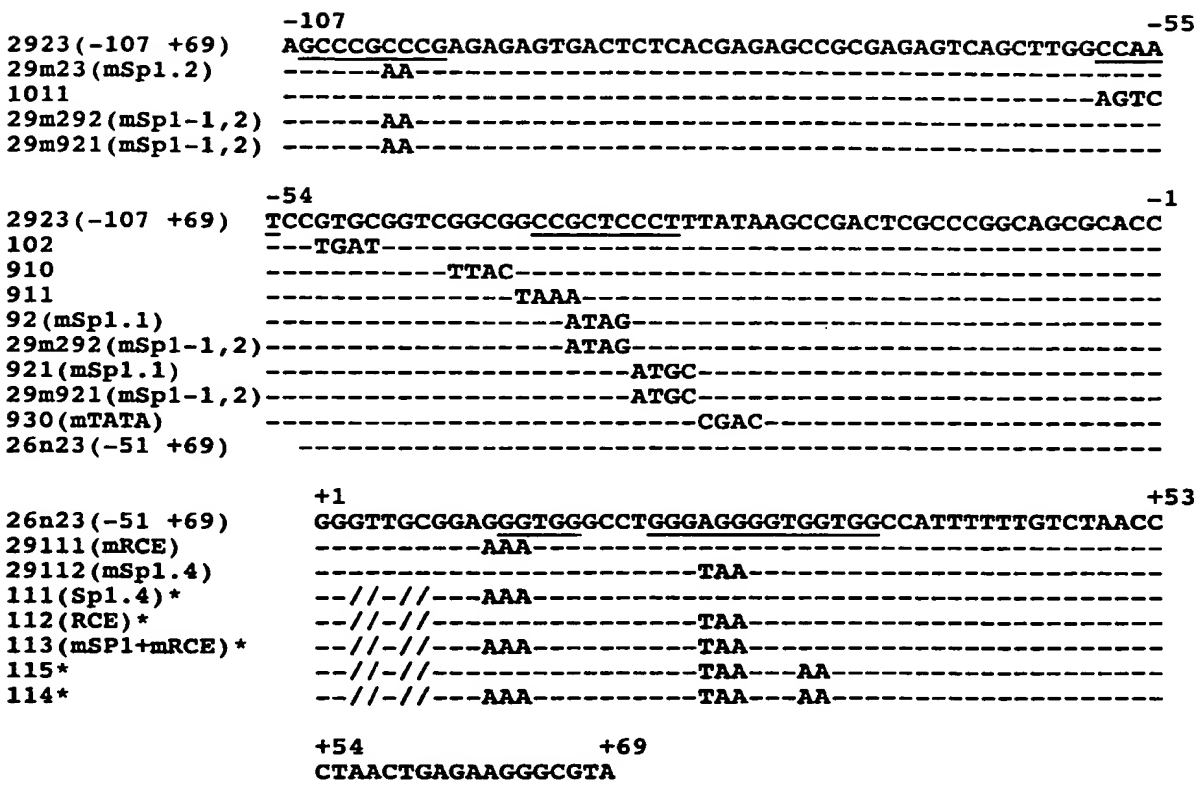

0
200
400
600
800
1000
1200
1400
1600
(Relative Luciferaes Activity)
K26(- ~4.5Kb +69)
E26(- ~2.5Kb +69)
hProm867(- 798 +69)
hProm697(-628 +69)
5F23(-471 +69)
hProm505(-463 +69)
hProm341(-2 72 +69)
2923(- 107 +69)
26n23(-51 +69)

Fig. 18



* Double mutagenesis by using pLh29m292 as template.

Fig. 19

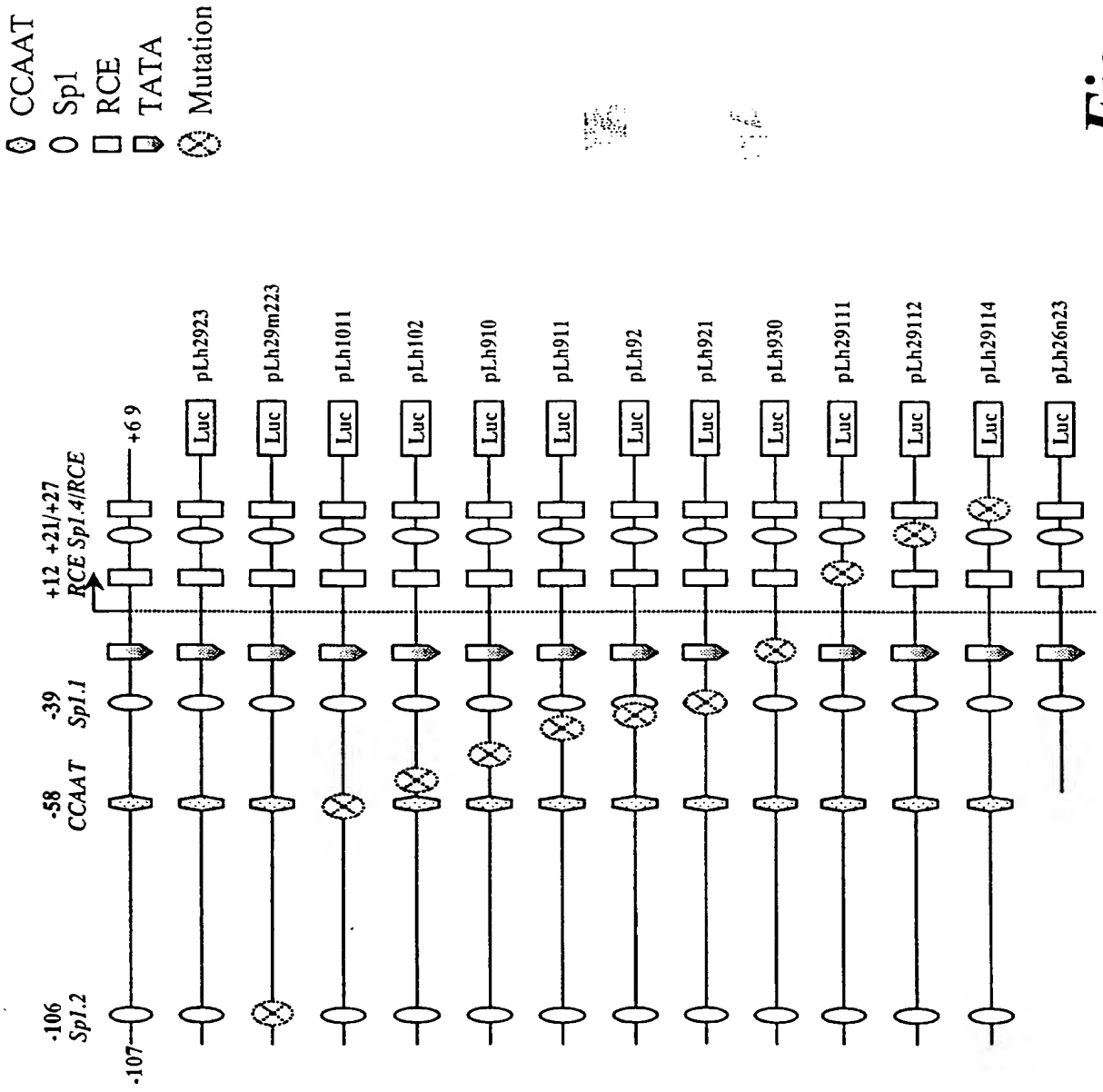

CCAAT
Sp1
RCE
TATA
Mutation
-107
-106
Sp1.2
-58
CCAAT
-39
Sp1.1
+12 +21/+27
RCE Sp1.4/RCE
+6 9
Luc
pLh2923
pLh29m223
pLh1011
pLh102
pLh910
pLh911
pLh92
pLh921
pLh930
pLh29111
pLh29112
pLh29114
pLh26n23

Fig. 20

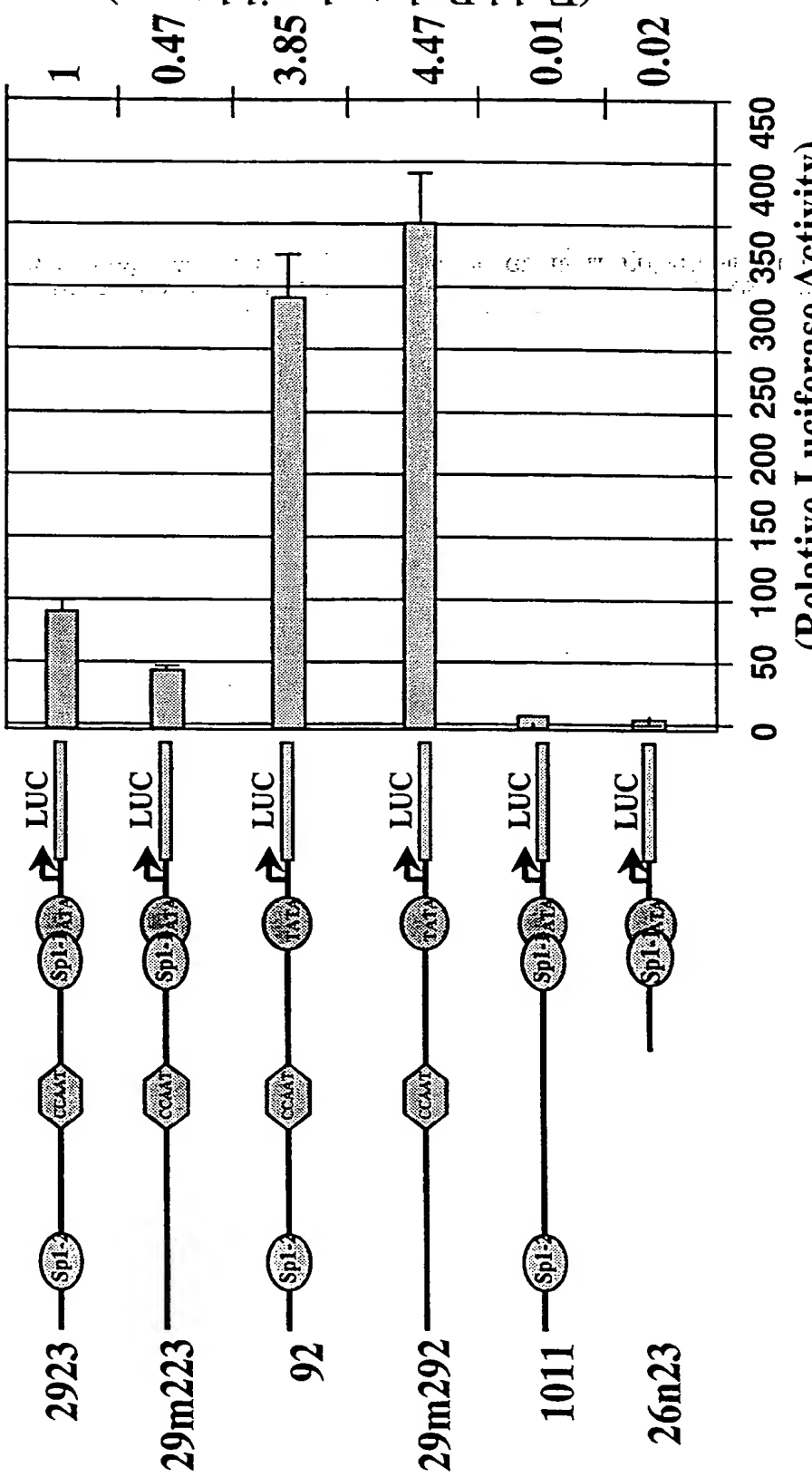

(Fold Related wild type)
1
0.47
3.85
4.47
0.01
0.02
0 50 100 150 200 250 300 350 400 450
(Relative Luciferase Activity)
LUC
Sp1
CCAAT
2923
29m223
92
29m292
1011
26n23

Fig. 21

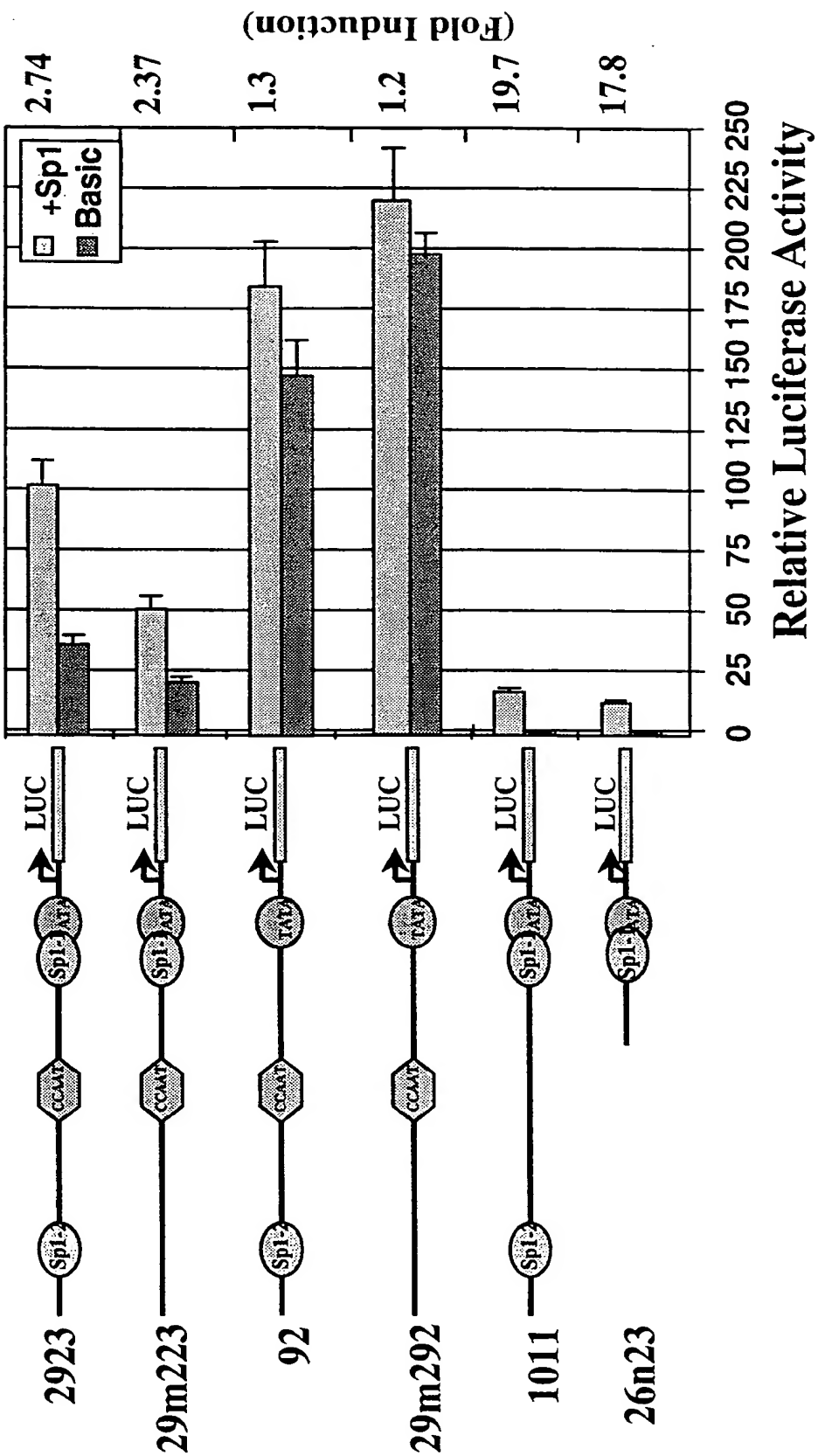

(Fold Induction)
2.74
2.37
1.3
1.2
19.7
17.8
+Sp1
Basic
0 25 50 75 100 125 150 175 200 225 250
Relative Luciferase Activity
LUC
Sp1
ATA
CCAAT
TATA
2923
29m223
92
29m292
1011
26n23

Fig. 22

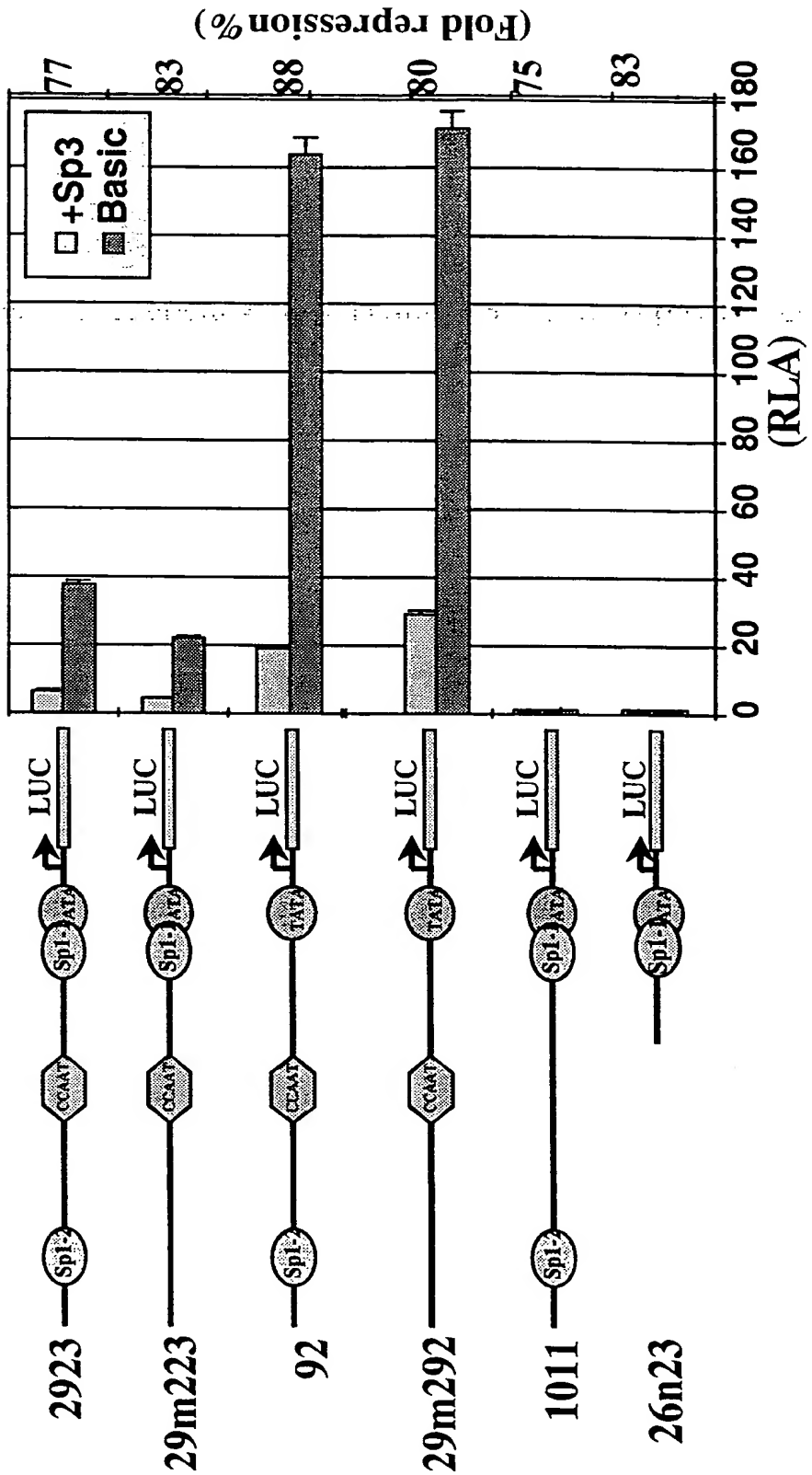

(Fold repression %)
77
83
88
80
75
83
+Sp3
Basic
0
20
40
60
80
100
120
140
160
180
(RLA)
LUC
Sp1
TATA
CCAAT
2923
29m223
92
29m292
1011
26n23

Fig. 23

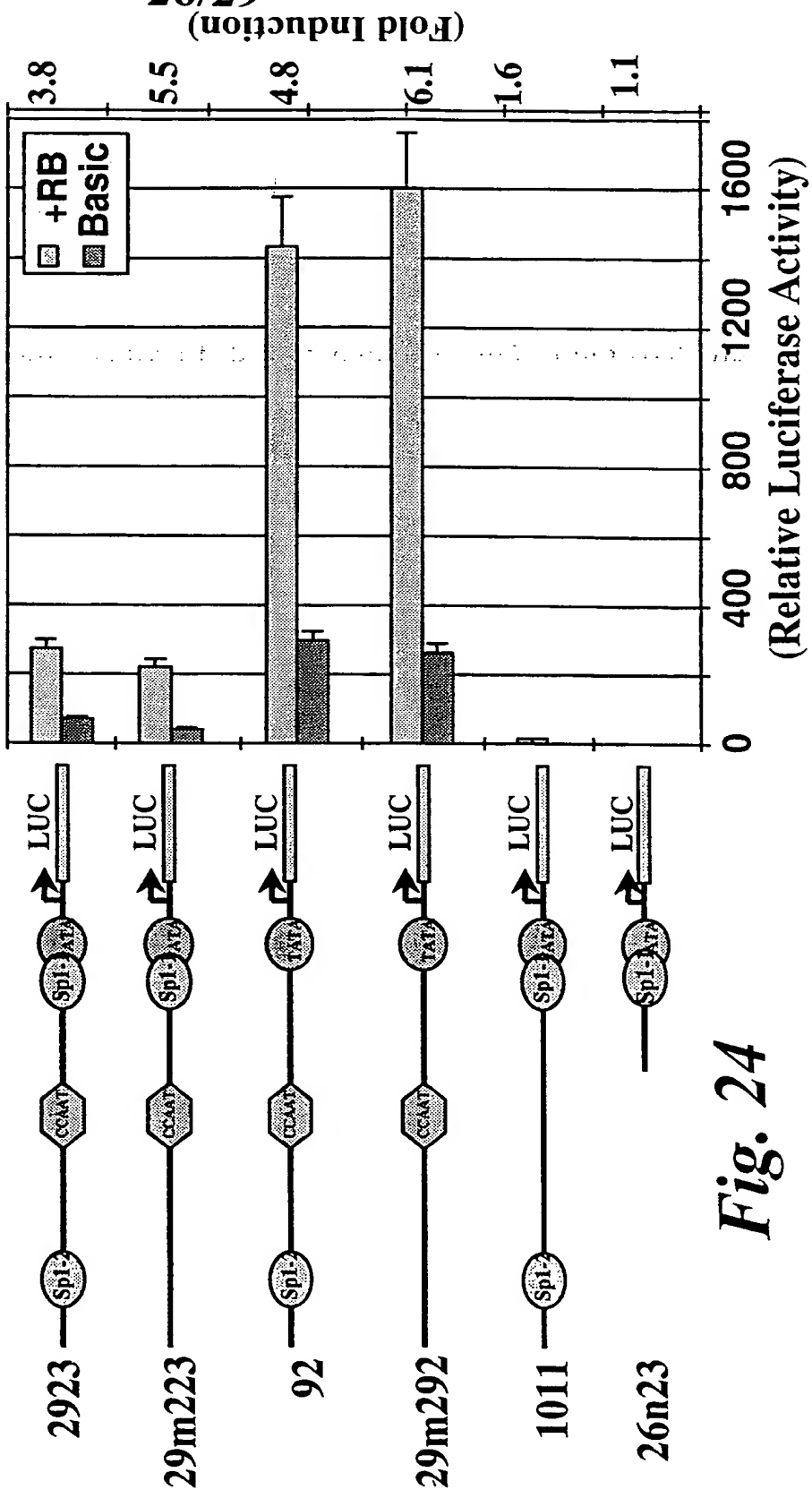

(Fold Induction)
3.8
5.5
4.8
6.1
1.6
1.1
+RB
Basic
0
400
800
1200
1600
(Relative Luciferase Activity)
LUC
Sp1
ATA
CCAAT
TATA
2923
29m223
92
29m292
1011
26n23

Fig. 24

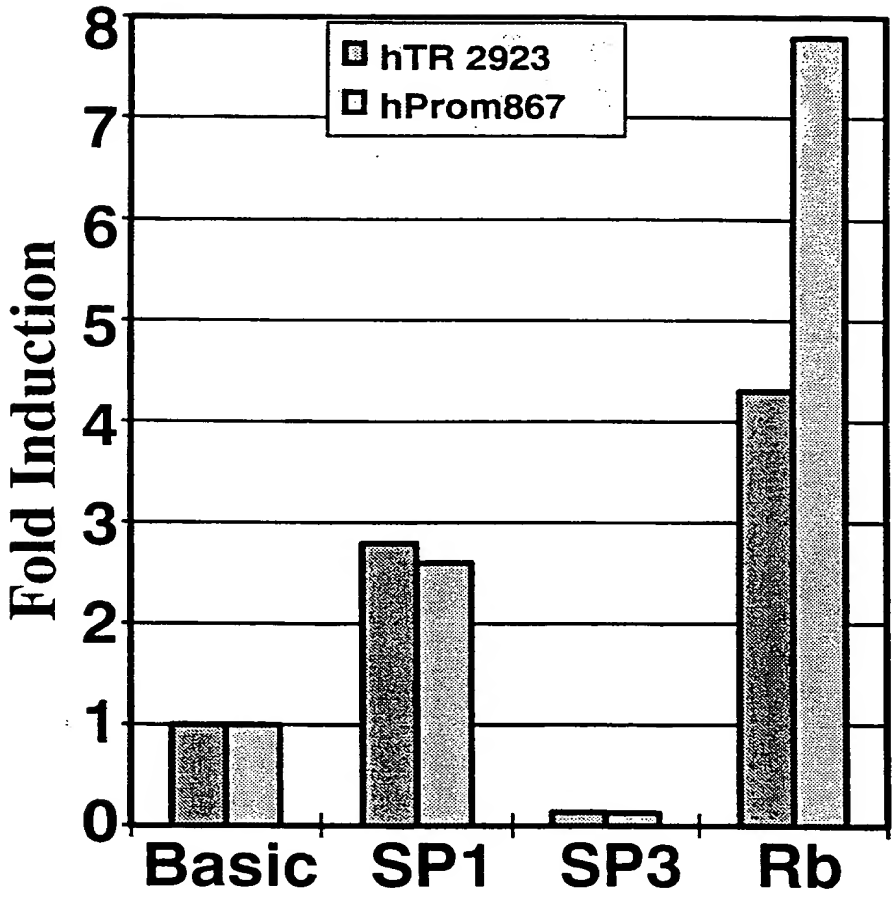
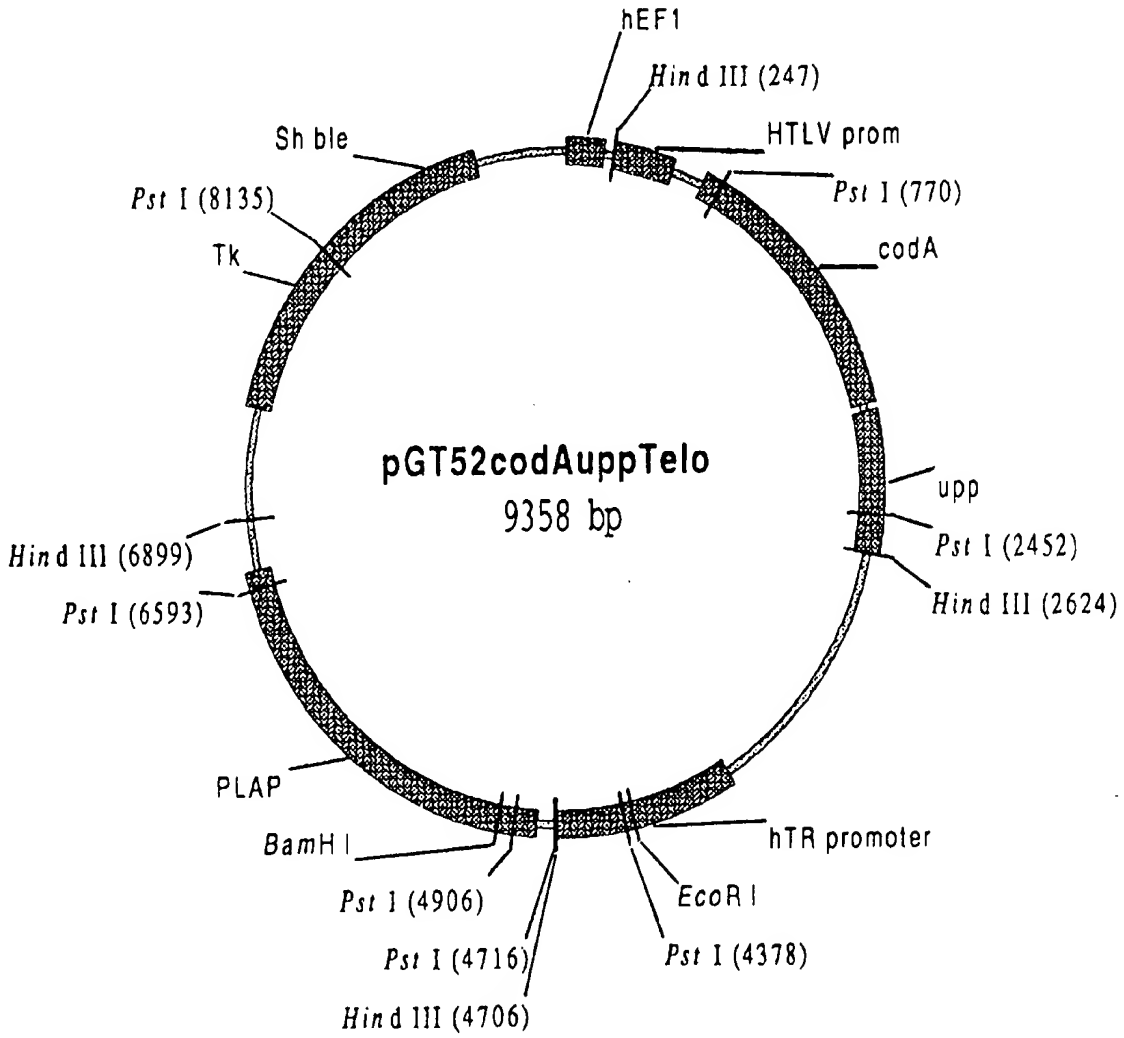

hTR 2923
hProm867
Fold Induction
0
1
2
3
4
5
6
7
8
Basic
SP1
SP3
Rb

Fig. 25



Feature Map
Misc. Feature (8 total)

hEF1
Start: 13 End: 200

HTLV prom
Start: 251 End: 534

codA
Start: 687 End: 1973
cytosine deaminase

upp
Start: 2001 End: 2629
uracil phosphoribosyltransferase

hTR promoter
Start: 3839 End: 4697

PLAP
Start: 4799 End: 6676
placental alkaline phosphatase

Tk
Start: 7383 End: 8501
HSV Thymidine kinase gene

Sh ble
Start: 8502 End: 8955
resistance to Zeocin

Fig. 26

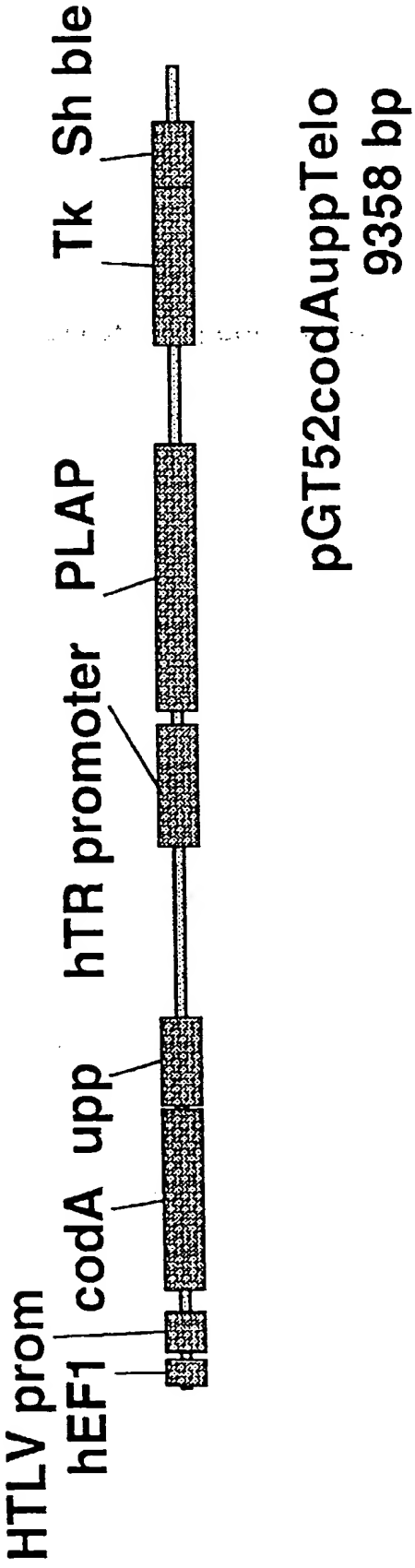

pGT52codAuppTelo
9358 bp
Sh ble
Tk
PLAP
hTR promoter
upp
codA
hEF1
HTLV prom

Fig. 27

PCT WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 6 :
C12N 15/11, 9/12, 15/85, C12Q 1/68

A3

(11) International Publication Number: **WO 99/38964**

(43) International Publication Date: 5 August 1999 (05.08.99)

(21) International Application Number: PCT/GB99/00308

(22) International Filing Date: 29 January 1999 (29.01.99)

(30) Priority Data:
9801902.9 29 January 1998 (29.01.98) GB

29 July 00/30 mos

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(72) Inventor; and

(75) Inventor/Applicant *(for US only)*: KEITH, William, Nicol [GB/GB]; CRC Dept. of Medical Oncology, University of Glasgow, CRC Beatson Laboratories, Garscube Estate, Switchback Road, Glasgow G61 1BD (GB).

(74) Agents: CRIPPS, Joanna, E. et al.; Mewburn Ellis, York House, 23 Kingsway, London WC2B 6HP (GB).

(81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

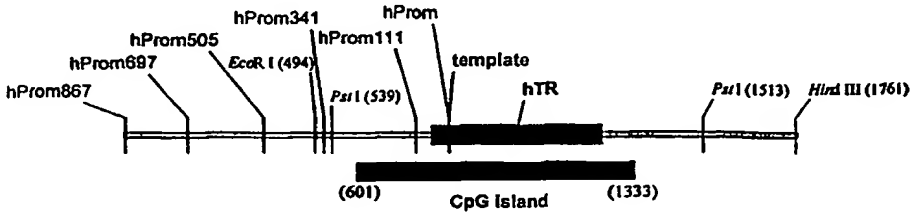
Published
With international search report.

(88) Date of publication of the international search report:
20 January 2000 (20.01.00)

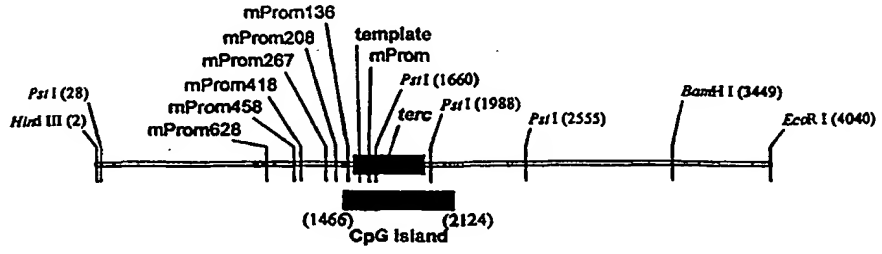
(54) **Title:** PROMOTER REGIONS OF THE MOUSE AND HUMAN TELOMERASE RNA COMPONENT GENES

(57) **Abstract**

The present invention relates to the identification of the genomic promoter region of the human and mouse telomerase RNA gene. Telomerase activity is necessary for the unrestricted proliferative capacity of many human cancers. It is proposed that mutation or dysregulation of the telomerase repression pathway may cause reactivation or upregulation of telomerase expression in cancer. The invention provides details of elements important for the regulation of telomerase RNA genes, including the Sp family of transcription factors. There is further provided methods for screening for elements having the ability for suppressing telomerase RNA gene promoter activity and use of such elements in the treatment of cancers. In addition, evidence is also provided for the development of new transcription based therapies for cancer and for genetic approaches to targeting therapeutic genes to cancer cells. Namely, (1) transcriptional repression and the disruption of signal transduction pathways regulating telomerase activation. (2) Tumour specific gene expression for genetic therapy via telomerase RNA gene promoters.



A



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INTERNATIONAL SEARCH REPORT

National Application No
PCT/GB 99/00308

A. CLASSIFICATION OF SUBJECT MATTER
IPC 6 C12N15/11 C12N9/12 C12N15/85 C12Q1/68

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 6 C12N C12Q

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 96 01835 A (ANDREWS WILLIAM H ;VILLEPONTEAU BRYANT (US); FUNK WALTER (US); FEN) 25 January 1996 (1996-01-25)	1-7,14, 15,17, 18, 23-28, 35,37,38
Y	page 24, line 13 -page 31, line 19 page 37, line 16 -page 43, line 13	15-21, 26-28, 30,31, 35,39-45
A	page 47, line 15 -page 55, line 19 page 65, line 9 -page 66, line 14	26-38

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[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

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Date of the actual completion of the international search	Date of mailing of the international search report
5 October 1999	19.10.1999

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL – 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Andres, S

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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category '	Citation of document, with indication.where appropnate. of the relevant passages	Relevant to claim No.
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A	the whole document	1-7
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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

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A	LANDBERG, G. ET AL.: "Telomerase activity is associated with cell cycle deregulation in human breast cancer" CANCER RESEARCH., vol. 57, 1 February 1997 (1997-02-01), pages 549-554, XP002117438 ISSN: 0008-5472	26-38
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P,X	ZHAO, J.-Q. ET AL.: "Cloning and characterization of human and mouse telomerase RNA gene promoter sequences." ONCOGENE, vol. 16, 1998, pages 1345-1350, XP002106808 ISSN: 0950-9232 the whole document	1-15,17, 18
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INTERNATIONAL SEARCH REPORT

International application No.
PCT/GB 99/00308

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

Although claims 44 and 45 are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

2. [X] Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:

see FURTHER INFORMATION sheet PCT/ISA/210

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

As a result of the prior review under R. 40.2(e) PCT, no additional fees are to be refunded.

1. [X] As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. [ ] As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[X] The additional search fees were accompanied by the applicant's protest.

[ ] No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.2

Present claims 26, 27 and 32 relate to a substance defined by reference to a desirable characteristic or property, namely its ability to modulate the activity of a TR promoter.
The claims cover all substances having this characteristic or property, whereas the application provides support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT for only a very limited number of such substances. In the present case, the claims so lack support, and the application so lacks disclosure, that a meaningful search over the whole of the claimed scope is impossible. Independent of the above reasoning, the claims also lack clarity (Article 6 PCT). An attempt is made to define the substance by reference to a result to be achieved. Again, this lack of clarity in the present case is such as to render a meaningful search over the whole of the claimed scope impossible. Consequently, the search has been carried out for those parts of the claims which appear to be clear, supported and disclosed, namely those parts relating to the specific modulators as defined in claims 28, 29 and 33.

The applicant's attention is drawn to the fact that claims, or parts of claims, relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. Claims: 2-7 (all totally) and claims 1,14,17-18, 37-38 (all partially)

   A nucleic acid comprising a promoter region of the human telomerase RNA gene, constructs, vectors and host cells containing it.

2. Claims: 8-13 (all totally) and claims 1,14,17-18, 37-38 (all partially)

   A nucleic acid comprising a promoter region of the mouse telomerase RNA gene, constructs, vectors and host cells containing it.

3. Claims: 16 (totally) and claims 15,17-22 (all partially)

   A nucleic acid construct comprising a telomerase RNA gene promoter linked to a cytotoxin, and its use in the treatment of cancer.

4. Claims: 23-36 (all totally) and claims 15, 17-22 (all partially)

   A method for screening for a substance being a modulator of the promoter of a telomerase RNA gene, the substances identified and their use in the treatment of cancer or the activation of telomerase.

5. Claims: 39-45

   A system for controlling neoplasia.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/GB 99/00308

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
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